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THE RÔLE OF THE HYPOPHYSIS IN CRANIAL OSTEOMYELITIS, PETROSITIS AND SINUS INFECTIONS

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It is generally conceded that osteomyelitis of the frontal bone arising from infections of the frontal sinus is one of the most devastating lesions encountered in surgical practise. In the same category we can place suppuration of the petrous portion of the temporal bone associated with mastoiditis. The clinical course of these lesions is sufficiently different from suppurative processes involving other skeletal foci, or even osteomyelitis of the cranium of traumatic origin to justify their consideration as distinct surgical entities.

Review of the minuter anatomy of these structures tends to explain the circumstances which ordinarily establish suppurative processes in these areas as very formidable surgical conditions. However, it is our purpose to present a new conception with regard to the incidence and influence of certain anatomical variations of known cause, which may, in some measure, meet the issue expressed by Furstenberg (1) that "none has offered a precise explanation of the influences which contribute to the development of cranial osteomyelitis."

There appear to be two important factors bearing on the development of cranial osteomyelitis of sinus origin and petrositis. These are, *A*). The blood-supply, *B*). the architecture of the skull.

A). The *Blood-Supply*. For the purpose of simplicity, it may be said that the outer table of the skull receives its blood-supply from superficial sources (the external periosteum), while the middle and inner tables receive their supply from within the cranium (dura). Likewise, the venous drainage of the outer table is in greater part outward to the superficial veins of the scalp, whereas those of the middle and inner tables drain inward, uniting with those of the dura to empty into the large sinuses of the dura.

Infections within the frontal sinus are thus readily provided with adequate

avenues of extension along and within the veins draining the middle and inner tables. It is in this manner that an epidural abscess appears early in the development of the disease, and the thrombophlebitis and periphlebitis readily assist in spreading the infection by direct continuity of their structures and by disturbing the blood-supply. The blood-supply thus becomes an important factor in explaining the spread of an osteomyelitic process arising from frontal sinus infections and is, undoubtedly, of major importance in explaining the extensive involvement of the hard, inner table and the epidural abscesses which are not unusual features of this disease.

The spread of the osteomyelitic process by means of the meningeal vessels may account for the appearance of the disease at distant foci about the skull. Bulson (2), quoting Guisez (3), who reviewed the work of Breschet (4) and of Schilling (5), states, " 'In all young subjects the infection appears to progress from one focus to another by virtue of the medullary richness of the diploetic element of bone.' " Luc (6) says that cranial osteomyelitis takes place only in those subjects presenting 'developmental anomaly'. He quotes Breschet, who observed that in general there is a greater development of the diploe in females. Thus, although earlier observers were aware of structural variations and their association with cranial osteomyelitis, they were not cognizant of the rôle played by the pituitary gland.

B). Architecture of the Skull. The bony texture of the skull is directly influenced by the activity of the anterior lobe of the hypophysis. This observation was brought to light in a study orginally based on 3000 skull roentgenograms at the Evans Memorial. The study (7) has since been extended to over 6000 cases. It has been found that various functional levels of the anterior lobe of the pituitary gland produce characteristic alterations in the structure of the skull. These changes have been classified into 4 groups or types.

Type 1. Overactivity of the anterior lobe produces an expansion of the trabecular network of bone. It may be likened to the effect of yeast on dough. The diploic table of the skull becomes expanded and more vascular, while the inner and outer tables are thinned. In consequence, those structures developing in diploe (the paranasal sinuses and mastoid cells) become enlarged. This mechanism finds its culmination in acromegaly (fig. 1). Here the vault is thick but light in structure, the diploe greatly expanded and the paranasal sinuses and mastoid cells hyperpneumatized. It is this type of case which also shows pneumatization of the petrous pyramid and apex (fig. 2). Lesser degrees of these changes are encountered more frequently than true acromegaly. The skulls of these patients, however, possess the same structural characteristics. The diploe is expanded and more vascular, and the paranasal sinuses and mastoids hyperpneumatized. This type of cranial dysplasia may vary in degree from slight departures from normal to a state properly designated as subacromegaly (fig. 3, 4).

Type 2. Post-adolescent or acquired deficiency of the anterior lobe results in skull changes which are just opposite to those produced by overactivity. Here we find an absence of diploic expansion. As a result, the sinuses and mastoid cells are poorly pneumatized. The skull is dense and hard, the sclerosis extending from the inner table into the diploe and from the sagittal plane laterally. The petrous pyramid is sclerotic and the tip non-pneumatized (fig. 5, 6). This type is frequently associated with obesity.

Type 3. Overactivity of the anterior lobe is frequently followed by hypofunctional involution. The skull roentgenograms of such patients present the typical features characteristic of each phase. The diploe is expanded, and the paranasal sinuses and mastoid cells hyperpneumatized. These are the residual evidences of the

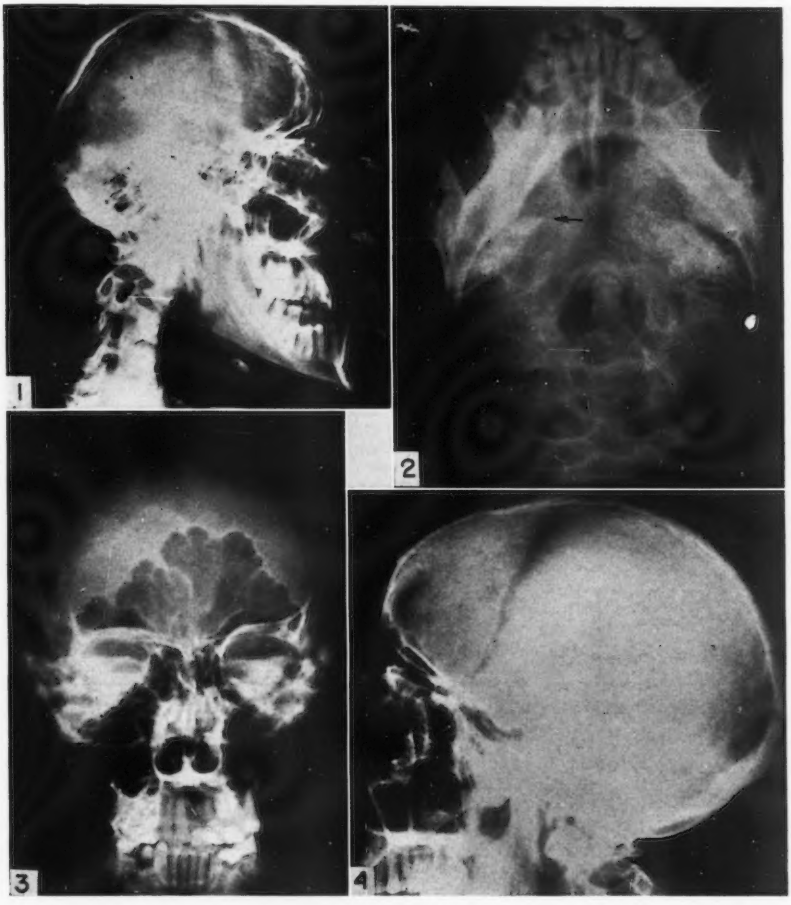

Fig. 1. Anterior lobe overactivity (acromegaly). The calvarium is thick, owing to expansion of the diploe. The outer and inner tables are comparatively thin. Note the marked degree of hyperpneumatization of the paranasal sinuses and mastoid cells. The bone is light and porous in structure as shown by the enlarged trabeculae, and in the prominent mandible. This is the prototype of the *type 1* skull.

Fig. 2. Base of skull showing petrous pyramids. From a case of anterior lobe overactivity. Note the well-formed pneumatic cells at the petrous apices (arrow).

Fig. 3. Anterior lobe overactivity. The general structural characteristics are the same as those observed in acromegaly, though to a lesser degree. Compare with fig. 1.

Fig. 4. Same case as fig. 3. Note the marked development of the frontal sinuses.

stage of overactivity. In addition, however, there is a secondary sclerosis beginning in the inner table and extending into the diploe. Frequently, there are patchy calcific deposits (enostoses) on the inner table. The changes are most marked in the frontal bone (fig. 7). The latter findings indicate the advent of a variable degree of anterior lobe failure.

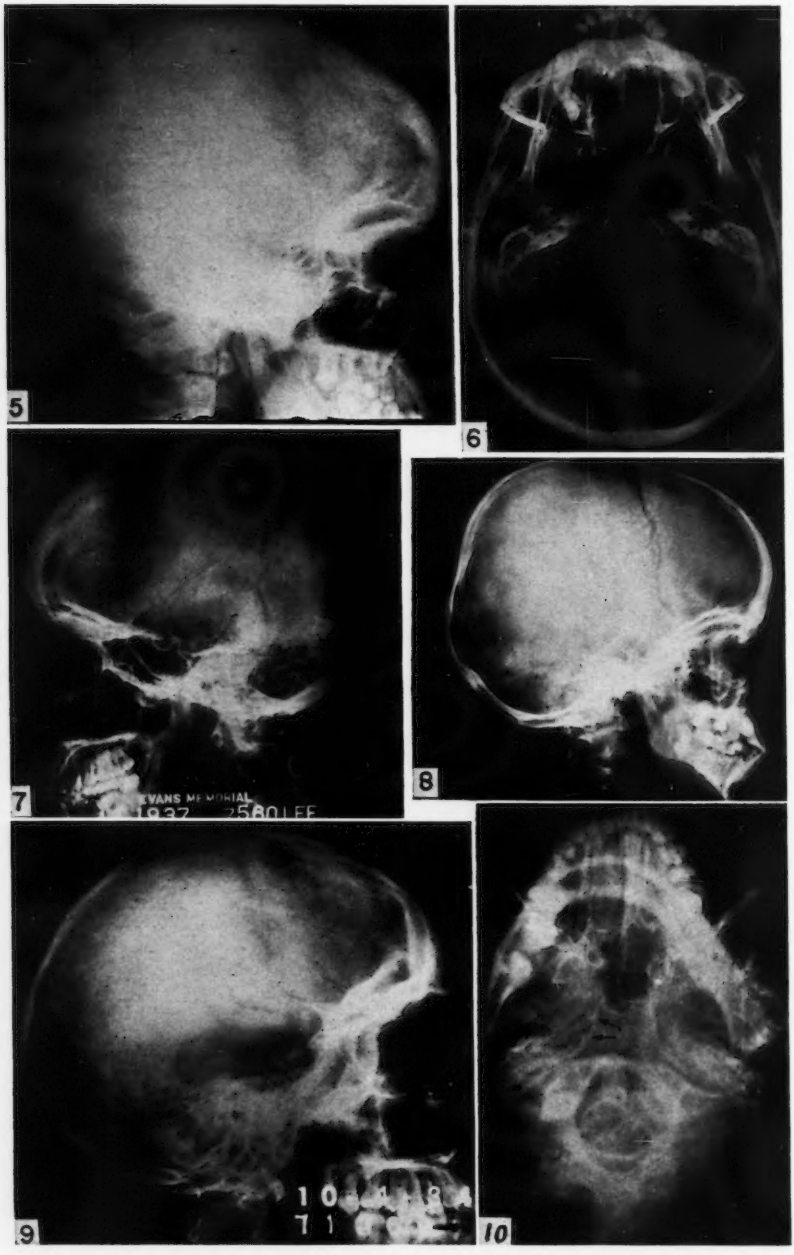

Fig. 5. *Type 2* skull. A case of anterior lobe failure associated with marked obesity. Note the small paranasal sinuses and mastoid cells, and the marked sclerosis of the vault which shows calcific deposits on the inner table. The bone is hard and dense.

Fig. 6. Base of skull showing petrous pyramids. From a case of anterior lobe failure. Note the fine cells in the pyramids and the non-pneumatized petrous apices (arrow).

Type 4. Congenital or pre-adolescent deficiency of the anterior lobe produces the type recognized as the 'pituitary dwarf'. The skull roentgenograms of these patients show a striking absence of diploic expansion, so that the outer and inner tables are in close approximation. As a result, the vault is thin, hard, dense and not vascular. The paranasal sinuses and mastoid cells are very poorly pneumatized, the petrous pyramids dense and cancellous. The frontal sinuses are frequently absent. The cranial sutures remain patent. The face and jaws are small and dentition is delayed (fig. 8).

These clinical observations were amply confirmed at a later date, by experimental studies on the white rat by Mortimer (8). Working with a series of 232 animals, he found that hypophysectomy resulted in a diminished vascularity of the bones of the cranium, inhibition of antero-posterior growth of the cranium and facial bones, hypoplosia of the diploe and frontal sinus homologue and retarded development of the teeth. Twenty-one hypophysectomized rats were then treated with somatotropic hormone, and it was found that the calvarial defects were largely corrected.

TABLE 1. INCIDENCE AND SEX DISTRIBUTION OF SKULL TYPES IN PITUITARY DYSFUNCTION IN 3000 CONSECUTIVE SKULL ROENTGENOGRAMS.

Type	Total	Males		Females		Total %
		No.	%	No.	%	
Normal	2457					
1	322	215	39.6	107	19.7	59.3
2	72	5	0.9	67	12.3	13.2
3	116	12	2.2	114	19.2	21.4
4	33	15	2.7	18	3.4	6.1

Study of the chimpanzee skull (pan panyscus and pan satyrus) has shown calvarial changes which parallel those observed in our clinical studies of human crania (9).

Of the 3000 cases which constituted the basis of the original study, 543 or 18.1 per cent were found to have roentgenological evidence of disturbed pituitary function. These are classified according to type and sex distribution in table 1, which shows that *type 1* occurs more commonly in the male, *types 2* and *3* in the female and *type 4* almost equally in both sexes.

The application of these observations to the present thesis is of more than mere academic interest. We have found that cranial osteomyelitis of sinus origin is usually found only in skulls having the structural characteristics of our *type 1* or *type 3* cases; that is, those having evidence of anterior lobe overactivity (fig. 9). The same is true of petrositis (fig. 10). Although it

Fig. 7. *Type 3* skull. Anterior lobe overactivity followed by hypofunctional involution. Female, aged 26, who started menstruating at 11 and attained full stature (5' 7") at 13, now has definite signs of hypopituitarism. Note the thick calvarium and extensive mastoid cells and paranasal sinuses which are residual evidences of anterior lobe overactivity. The marked degree of sclerosis of the frontal bone indicates the advent of anterior lobe deficiency. This case combines the essential features of *types 1* and *2*. Compare with fig. 1 and 5.

Fig. 8. *Type 4* skull. Congenital or pre-adolescent deficiency ('pituitary dwarf'). Patient aged 29. Note the thin calvarium owing to poorly developed diploe. The frontal sinuses are absent, and the other paranasal sinuses and mastoid cells poorly pneumatized. The bone is dense, hard and heavy.

Fig. 9. Case of osteomyelitis of the frontal bone arising from frontal sinus infection, running a rapid, fulminating course terminating in death. Note the extent of the sphenoid sinus and mastoid cells. The general characteristics are those of the *type 1* skull.

Fig. 10. Case of petrositis following simple mastoidectomy in a patient aged 14. Pneumatization of the petrous apex typical of the *type 1* skull is seen on the un-involved right side (arrow). Diseased apex on left.

has been recognized by other observers (10, 11, 12) that pneumatization of the petrous pyramid is a variable factor, we are of the opinion that its structure is dependent upon the functional activity of the pituitary gland.

Our observations have led us to the conclusion that the structural differences between skulls corresponding to the various phases of pituitary activity

TABLE 2. INCIDENCE OF SINUS INFECTION IN 1286 CONSECUTIVE CASES.

Skull Type	Number	Sinus Infection	%
Pituitary Dysfunction	472	189	40
Normal	814	272	23

are sufficiently great to allow the development of frontal osteomyelitis or suppuration of the petrous apex in one individual and not in another. It would appear that the rarefied, spongy and vascular bone in the one case and the sclerotic, dense bone in the other, are determining factors.

We have observed also, that persistent drainage following simple mastoidectomy occurs most often in cases belonging to the *type 1* group. Here, again the loose bony structure is probably less resistant to infection. Moreover, the mastoid is more extensively pneumatized, almost invariably having zygomatic cells which, though involved in the suppurative process, may be hidden by a ridge of bone and be overlooked at operation.

DISCUSSION

It thus becomes of practical importance to recognize skull types in the treatment of infections of the paranasal sinuses and mastoids. Patients having skulls of the *type 1* or *type 3* group not only merit more careful surgical

TABLE 3. RELATION OF SKULL TYPE AND SINUS INFECTION IN 472 CASES OF PITUITARY DYSFUNCTION.

Type of Skull	Sinus Infection	%
1 and *3*	136	71.9
2 and *4*	53	28.1

technic but also closer post-operative observation. It is in such patients that frontal osteomyelitis or suppuration of the petrous apex may develop. Our own series of cases of this kind being rather small (9), we reviewed a series of 14 unselected cases at the Massachusetts Eye and Ear Infirmary, through the kindness of Dr. A. S. MacMillan. Every one of these could be classified as belonging to *type 1* or *type 3*. Some had a definite acromegalic appearance.

The incidence of infection in these cases is also of interest. Of 1286 consecutive cases recently examined, in which a complete series of skull and sinus films were available, 472 showed evidence of disturbed pituitary function and 814 were normal. As shown in table 2, there was roentgenological evidence of sinus infection in 23 per cent of the normal skull cases and in 40 per cent of those showing pituitary dysfunction. This observation stresses the importance of ascertaining the endocrine factor in the treatment of these conditions.

Allergic ethmoiditis seems to bear a definite relation to pituitary dysfunction. Studies along these lines are now in progress. It is further interesting to note in table 3 that of the cases of pituitary dysfunction having sinus infections, 71.9 per cent were of *type 1* and *type 3* combined. These are the patients showing skull changes of overactivity. *Types 2* and *4* (showing the changes of pituitary deficiency) combined to make a total of 28.1 per cent.

In considering the possible mechanism responsible for the susceptibility of certain types of skull to osteomyelitis or petrositis it becomes apparent that texture and vascularity of bone and amount of calcium are of major importance. It is our belief that even the initial infection may be shown to bear definite relation to the activity of the pituitary gland.

SUMMARY

The architecture of the skull is influenced by the activity of the anterior lobe of the hypophysis. These changes, which are revealed by the roentgenogram, are classified at *types 1, 2, 3* and *4*. Cranial osteomyelitis of sinus origin and suppuration of the petrous apex usually occur in *types 1* or *3*. These types are associated with hyperactivity of the anterior lobe.

The incidence of sinus infection is greater in those having evidence of pituitary dysfunction than it is in people with normal skulls, and of the former it is more common in *types 1* and *3* than it is in *types 2* and *4*.

Our studies indicate the necessity of considering skull types and structure in the treatment of infections of the paranasal sinuses and mastoids, and suggest that the endocrine factor plays an important rôle in the development and progress of these conditions.

REFERENCES

1. Furstenburg, A. C.: Ann. Otol., Rhin. & Laryng. **40**:996. 1931.
2. Bulson, A. E.: Trans. Am. Ophth. & Otol. P. 102. 1925.
3. Guisez, J.: Ann. d. mal de l'oreille, du larynx. **32**:600. 1906.
4. Breschet, G.: Essai sur les veines de rachis, recherches sur la formation du cal; considerations et observations anatomiques et pathologiques. (Thesis) Paris, 1819.
5. Schilling, V.: Ztschr. f. Ohrenhk. **48**:52. 1904.
6. Luc, H.: Ann. de mal de l'oreille, du larynx **35**:236. 1909.
7. Mortimer, H., G. Levene and A. W. Rowe: Radiology **29**. No. 2:135. 1937, *ibid.*, No. 3:279. 1937.
8. Mortimer, H.: Radiology **28**:5. 1937.
9. Coolidge, H. J., L. Wyman and G. Levene: Exhibit A.A.A.S., Cambridge, 1933.
10. Eagleton, W. P.: Arch. Surg. **20**:386. 1930.
11. Kopetzky, S. J.: Arch. Otolaryng. **18**:47. 1933.
12. Taylor, H. K.: Arch. Otolaryng. **18**:458. 1933.

THE RELATION OF THE IODINE TOLERANCE TO THYROID FUNCTION

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In a previous paper (1) we described an iodine tolerance test whereby the relative rate of disappearance from the blood stream of intravenously injected iodine might be expressed. The results of the application of the test in a small series of cases showed that, in patients with clinical and histological signs of thyrotoxicosis, the injected iodine disappeared from the blood stream more rapidly than in persons with clinically normal thyroid glands. Also, in patients with hypothyroidism, the average quantity of iodine in the blood 6 hours after its injection was greater than normal. It was intimated that such a method might provide evidence of diagnostic importance in cases of doubtful thyroid disease.

The present report is based upon a more extensive series of cases, but in the main the deductions expressed in the first communication have been reaffirmed. In addition, experience with the iodine tolerance test has led to the assumption that a more comprehensive understanding of endocrine disorders may be acquired by studies of the iodine metabolism.

The intravenous iodine tolerance test. Iodine tolerance tests have, as their basis, variations in the affinity of the thyroid gland for iodine. In accordance with this principle, a modification of a method described by Elmer (2) was devised for determining, in a specified time, the rate and completeness of disappearance from the blood stream of a calculated amount of iodine in the form of Lugol's solution, following its intravenous injection. The results are expressed in terms of percentage of the injected iodine remaining in the blood stream at intervals after the injection. While these values represent the relative rather than the absolute iodine concentrations, they do provide an indication of the rate and the completeness with which the injected iodine disappears from the circulation during the course of the experiment.

Theoretically at least, this procedure should supply evidence concerning the state of functional activity of the thyroid gland. Marine and his associates (3, 4) demonstrated, by means of perfusion, experiments that the thyroid had a marked and selective affinity for iodine. The absorption of potassium iodide by the gland took place immediately and hyperplastic glands, with a low iodine content, removed the greatest quantities of iodine from the perfusion fluid. Moreover, the iodine taken up by the thyroid could not be removed by subsequent washing, indicating that it was bound and concentrated in the thy-

roid. Apparently this gland possesses the only tissue in the body capable of retaining iodine. Even traces could not be detected in any organs, with the exception of the thyroid, after they were freed of blood. There is, therefore, experimental precedent for the intravenous iodine tolerance test.

The technique of the intravenous iodine tolerance test. Space will not be utilized for a detailed account of the intravenous iodine tolerance test, which has been described elsewhere (1, 5), except to mention that the patient, fasting, receives an intravenous injection of Lugol's solution containing 250 γ of iodine per kg. of body weight. Samples of venous blood are withdrawn before the injection, and at intervals of 5 minutes, ½ hr., 2 hr., 4 hr., and 6 hr. afterward. Estimations of the iodine in these samples of blood were performed by a modification of the method described by Perkin (6).

TABLE 1. RESULTS OF THE IODINE TOLERANCE TEST IN PERSONS WITHOUT FUNCTIONAL ABNORMALITY OF THE THYROID GLAND. CONTROL CASES.

Case and Sex	Age Yr.	B.M.R. deviation %	Percentage of injected I remaining in blood stream				Iodine excreted in urine, γ	Diagnosis
			½ hr.	2 hr.	4 hr.	6 hr.		
1F	27	+10	64	18	8	5	992	Anxiety neurosis
2F	48	+2	29	25	—	5	533	Simple obesity
3M	54	+7	82	36	27	9	1362	Chronic bronchitis
4F	22	0	67	54	25	9	176	Normal
5F	23	−4	61	39	22	9	314	Normal
6F	38	+9	52	34	26	9	818	Post-op. removal of coccyx
7M	21	−2	44	18	14	9	334	Familial periodic paralysis
8F	14	−4	70	31	20	11	198	Scoliosis
9F	15	−1	75	55	35	14	761	Post-appendectomy
10F	30	+9	59	—	22	15	?	Neurosis
11M	45	+4	74	49	27	16	330	Chronic arthritis
12M	58	+9	77	60	32	18	947	Chronic bronchitis
13M	25	+7	63	47	38	18	444	Varicose ulcer
14M	57	+4	84	65	45	19	1396	Cardiovascular degeneration
15M	31	−7	64	54	27	20	265	Normal
16M	33	−1	54	38	24	20	324	Sciatic scoliosis
17F	42	+3	79	63	52	21	342	Dysmenorrhoea
18F	31	−6	86	58	40	22	1142	Mental deficiency
19M	16	+5	63	47	36	23	405	Pseudo-ganglion of foot
20M	54	+2	49	26	23	23	309	Peptic ulcer
21M	36	−2	85	62	50	31	533	Peptic ulcer
22M	29	+8	92	71	57	32	317	Brain tumor?
23M	24	−1	66	38	38	32	44	Bronchial asthma
24M	22	−1	71	49	41	38	400	Bronchitis
Average		+2	67	45	32	18	552	

The iodine content of the blood specimen secured 5 minutes after the injection, minus that of the preliminary control sample, is regarded as representing the maximum increment caused by the injected iodine and it is, consequently, recorded as 100 per cent. With this value as a basis, the findings for the other samples are expressed accordingly. A curve of the percentages of the injected iodine remaining in the peripheral blood stream at the various test-periods may be plotted. This procedure is an adaptation of the principle employed in the bilirubin excretion test of liver function of von Bergmann (7).

TABLE 2. RESULTS OF THE IODINE TOLERANCE TEST IN PATIENTS WITH THYROTOXICOSIS.

Case and Sex	Age Yr.	B.M.R. deviation %	Percentage of injected I remaining in blood stream				Iodine excreted in urine, γ	Diagnosis
			½ hr.	2 hr.	4 hr.	6 hr.		
With Hypermetabolism								
*25*F	30	+35	79	56	0	0	178	Graves's disease
*26*F	33	+41	54	22	0	0	277	Graves's disease
*27*F	66	+15	34	34	2	0	30	Nodular goiter
*28*F	62	+32	29	10	3	0	477	Graves's disease
*29*F	39	+39	71	47	6	0	515	Graves's disease
*30*F	45	+66	52	41	7	0	592	Graves's disease
*31*M	44	+32	36	27	7	0	289	Graves's disease
*32*F	36	?	33	16	8	0	95	Graves's disease
*33*M	49	+46	53	33	10	0	84	Nodular goiter
*34*F	23	+31	53	20	15	0	212	Graves's disease
*35*F	60	+35	59	42	20	0	205	Graves's disease
*36*F	47	+22	60	38	23	0	?	Nodular goiter
*37*M	47	+40	58	56	44	0	33	Graves's disease
*38*F	62	+74	74	57	57	1	114	Graves's disease
*39*M	33	+25	39	5	3	2	1066	Recurrent Graves's disease
*40*F	68	+40	45	33	25	3	1028	Nodular goiter, congestive heart failure
*41*F	59	+73	84	52	4	4	635	Graves's disease
*42*F	43	+33	40	31	31	4	457	Nodular goiter
*43*F	43	+30	56	51	24	4	82	Graves's disease
*44*F	48	+22	29	18	18	4	337	Graves's disease
*45*F	51	+28	66	56	25	5	121	Graves's disease
*46*F	61	+39	48	38	33	5	80	Nodular goiter
*47*F	28	+30	81	61	29	7	696	Thyrotoxicosis
*48*F	34	+36	31	25	17	8	723	Recurrent Graves's disease
Without Hypermetabolism								
*49*M	37	+14	84	57	46	5	212	Recurrent Graves's disease
*50*M	31	+7	70	39	23	0	201	Nodular goiter
*51*M	32	+1	64	29	29	0	1734	Hyperthyroidism
Average		+34	55	36	19	2	403	

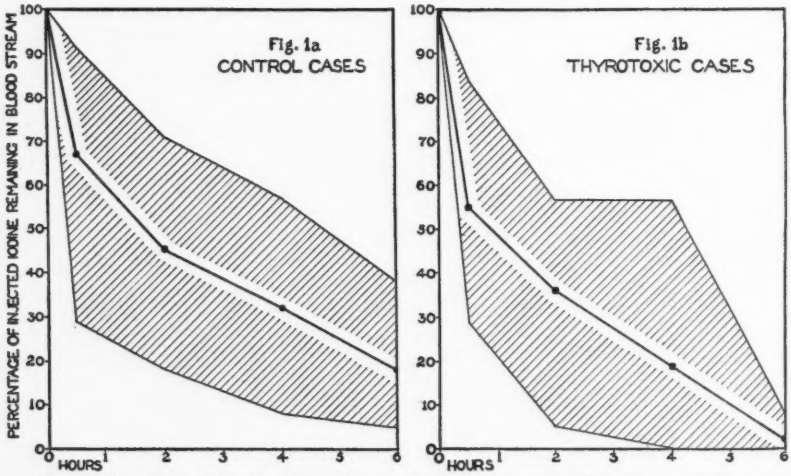

Fig. 1*a*. Showing the limits of variation of the results of the intravenous iodine tolerance test in persons without abnormality of the thyroid gland.

Fig. 1*b*. Showing the limits of variation of the results of the intravenous iodine tolerance test in patients with thyrotoxicosis.

TABLE 3. THE EFFECT OF THYROIDECTOMY UPON THE RESULTS OF THE IODINE TOLERANCE TEST.

Case and Sex	Age Yr.	B.M.R. deviation %	Percentage of injected I remaining in blood stream				Iodine excreted in urine, γ	
			½ hr.	2 hr.	4 hr.	6 hr.		
*25*F	30	+35	79	56	0	0	178	Pre-operatively
		− 8	88	53	44	35	459	Postoperatively
*26*F	33	+41	54	22	0	0	277	Pre-operatively
		− 6	83	71	54	38	56	Postoperatively
*30*F	45	+66	52	41	7	0	592	Pre-operatively
		−10	83	43	28	14	1357	Postoperatively
*33*M	49	+46	53	33	10	0	84	Pre-operatively
		− 6	65	47	41	41	45	Postoperatively
*36*F	48	+22	60	38	23	0	?	Pre-operatively
		+29	100	82	66	33	618	Postoperatively
*39*M	33	+25	39	5	3	2	1066	Pre-operatively
		−14	40	19	15	10	1058	Postoperatively
*41*F	59	+73	84	52	4	4	635	Pre-operatively
		+12	98	75	42	28	273	Postoperatively
*42*F	43	+33	40	31	31	4	457	Pre-operatively
		−17	77	64	34	13	1184	Postoperatively
*44*F	48	+22	29	18	18	4	337	Pre-operatively
		− 2	58	58	34	34	846	Postoperatively
*45*F	51	+28	66	56	25	5	121	Pre-operatively
		− 3	71	57	48	14	389	Postoperatively
Average		+39	56	35	12	2	375	Pre-operatively
		− 3	76	57	41	26	629	Postoperatively

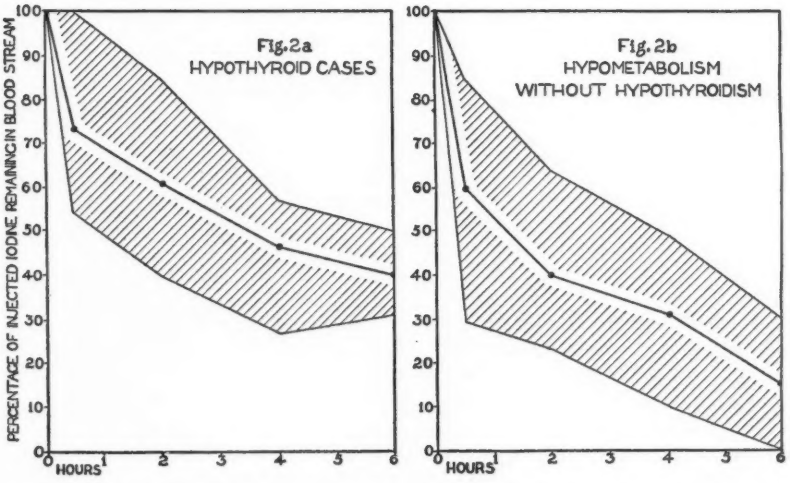

Fig. 2*a*. Showing the limits of variation of the results of the intravenous iodine tolerance test in patients with hypothyroidism.
Fig. 2*b*. Showing the limits of variation of the results of the intravenous iodine tolerance test in patients with hypometabolism without the clinical manifestations of hypothyroidism.

OBSERVATIONS

The results of 91 intravenous iodine tolerance tests performed on 80 patients are recorded in this paper. The individuals tested included 24 hospital patients without obvious thyroid disease; 27 patients with thyrotoxicosis, 10 of whom were tested after thyroidectomy as well as before; 5 patients with non-toxic goiters; 6 patients with hypothyroidism and 18 persons with hypometabolism without the clinical signs of hypothyroidism. Some of the observations were included in the previous report (1).

Control Cases. In order to determine the response of persons apparently with functionally normal thyroid glands, a number of subjects were selected for the test from the general medical and surgical wards of the Victoria Hospital. While these individuals cannot be regarded as 'normal' in the ordinary sense of the term, they may be considered representative of the population of hospital patients without functional abnormality of the thyroid gland. All had basal metabolic rates within the generally accepted range of the normal, i.e., —10 per cent to +15 per cent.

The results obtained in 24 such cases are shown in table 1. Special reference is directed to the percentage of the iodine remaining in the blood stream 6 hours after its injection. As may be seen in figure 1*a*, which represents the limits of variation in this control series, there is a considerable spread of the values, but in 18 of the 24 cases the 6-hour retentions fell within a range of 9 to 23 per cent. In 2 instances, only 5 per cent of the injected iodine remained in the circulation after 6 hours and, in 4 cases, more than 23 per cent. The average for the whole group at the 6-hour period was 18 per cent.

Thyrotoxic cases. Quite a different impression is conveyed by table 2 and figure 1*b*, which depict the findings in 27 patients with clinical and histological evidences of thyrotoxicosis due to Graves's disease, or to nodular goiter with hyperthyroidism. In the majority of these patients, the B.M.R. was excessive. *Case 32* was suffering from a degree of thyrotoxicosis which precluded a satisfactory estimation of the B.M.R.

It is obvious that the injected iodine had disappeared from the blood stream more rapidly in the patients of this group than in those of the control series. In 13 of the 27 cases, not any of the injected iodine remained in the peripheral blood at the end of 6 hours. The highest 6-hour figure for the whole group was only 8 per cent, in a patient with a postoperative recurrence of Graves's disease. The general average for the thyrotoxic patients was 2 per cent. No essential difference was noted between the response of patients with Graves's disease and those with toxic nodular goiter.

Estimations of the amounts of iodine excreted in the urine during the test periods were performed. Although there were wide fluctuations in the quantities of iodine eliminated, as shown by the tables, the average output was less for the thyrotoxic group than for the controls, probably indicating that the more rapid disappearance of the iodine from the circulation in the thyrotoxic patients was not due to loss of iodine through the kidneys but rather to fixation by the hyperplastic thyroid glands.

The effect of thyroidectomy. It is interesting to note the effect of subtotal thyroidectomy upon the results of the iodine tolerance test in patients with

previous thyrotoxicosis. The differences in the rate of clearance of the blood of the iodine before and after that operation, performed on 10 patients, are shown in table 3 and figure 3. In every case the injected iodine left the circulation much more slowly after thyroidectomy than before.

Non-toxic Goiter. The results of the iodine tolerance test in 5 patients with non-toxic goiters are presented in table 4. The findings were essentially the same as those obtained in the control series of cases (table 1).

TABLE 4. RESULTS OF THE IODINE TOLERANCE TEST IN PATIENTS WITH NON-TOXIC GOITERS.

Case and Sex	Age Yr.	B.M.R. deviation %	Percentage of injected I remaining in blood stream				Iodine excreted in urine, γ	Diagnosis
			½ hr.	2 hr.	4 hr.	6 hr.		
*52*F	21	+8	65	35	21	9	108	Colloid goiter
*53*M	54	−1	57	36	29	14	571	Nodular goiter Diabetes mellitus (mild)
*54*M	54	−1	88	36	31	15	1580	Diffuse colloid goiter
*55*F	68	+13	62	35	30	24	964	Nodular goiter Diabetes mellitus
*56*F	38	−4	86	73	47	26	307	Colloid adenoma
Average		+3	72	43	32	18	706	

Hypothyroidism. The results in a small group of 6 patients with hypothyroidism are shown in table 5 and figure 2*a*. The diagnosis of hypothyroidism was based upon the clinical features, a lowering of the B.M.R. and an increase of the blood cholesterol. One patient (*62*) showed the clinical manifestations of hypothyroidism but without hypometabolism.

TABLE 5. RESULTS OF THE IODINE TOLERANCE TEST IN PATIENTS WITH HYPOTHYROIDISM.

Case and Sex	Age Yr.	B.M.R. deviation %	Percentage of injected I remaining in blood stream				Iodine excreted in urine, γ	Diagnosis
			½ hr.	2 hr.	4 hr.	6 hr.		
Hypothyroidism								
*57*F	65	−37	71	57	57	50	38	Myxedema
*58*F	56	−25	65	50	48	46	15	Hypothyroidism
*59*M	36	−24	74	67	41	41	1574	Hypothyroidism
*60*F	30	−20	54	47	44	33	?	Hypothyroidism
*61*F	25	−25	100	85	38	31	1049	Hypothyroidism
Clinical Signs of Hypothyroidism Without Hypometabolism								
*62*F	34	− 5	80	67	46	33	829	Hypothyroidism(?)
Average		−23	74	62	46	39	701	
The Effect of Thyroid Medication								
*57*F	65	−37	71	57	57	50	38	Before treatment
		− 8	77	40	27	0	152	After treatment

In 3 of the 6 cases, the percentage of iodine remaining in the blood at the end of the iodine tolerance test was considerably in excess of the values ex-

hibited by the control group of patients in table 1. There was an average of 39 per cent in the hypothyroid patients as compared with 18 per cent in the controls.

The effect of thyroid medication in one case of typical myxoedema was rather striking, as depicted in table 5*c*. This patient (*57*) showed a six-hour retention of 50 per cent of the injected iodine in the blood before the institution of treatment with desiccated thyroid. After the B.M.R. had arisen from —37 per cent to —8 per cent with an accompanying marked clinical improvement as a result of the treatment, all of the injected iodine had disappeared in 6 hours.

Hypometabolism without clinical hypothyroidism. Table 6 and figure 2*b* show the results obtained in a heterogeneous group of 18 patients with but one

TABLE 6. RESULTS OF THE IODINE TOLERANCE TEST IN PERSONS WITH HYPOMETABOLISM WITHOUT CLINICAL SIGNS OF HYPOTHYROIDISM.

Case and Sex	Age Yr.	B.M.R. deviation %	Percentage of injected I remaining in blood stream				Iodine excreted in urine, γ	Diagnosis
			½ hr.	2 hr.	4 hr.	6 hr.		
*63*F	43	−30	60	30	30	0	42	Exhaustion syndrome
*64*F	41	−19	65	46	30	0	296	Peripheral neuritis
*65*M	33	−13	29	23	10	3	132	No diagnosis
*66*F	38	−23	57	35	21	7	320	Dysmenorrhoea
*67*M	33	−38	31	23	—	8	656	Polyglandular syndrome
*68*F	36	−16	44	40	23	9	1015	Pituitary dysfunction?
*69*M	42	−32	61	47	29	9	1557	Postinfluenza
*70*M	39	−15	44	—	33	11	514	Exhaustion syndrome
*71*M	30	−15	71	28	21	14	698	Chr. (healed) Pulmonary Tb.
*72*F	46	−22	60	55	28	15	124	Erosion of cervix
*73*M	46	−16	59	35	29	18	431	Chronic sinusitis
*74*M	41	−31	80	40	37	20	1306	Pneumonia (convalescent)
*75*M	24	−15	63	31	31	21	71	Psychoneurosis
*76*F	15	−26	55	46	35	22	177	Postoperative adhesions
*77*F	29	−18	80	64	49	23	914	Pleurisy
*78*M	24	−21	85	33	29	23	163	Postinfluenza
*79*F	34	−24	59	40	40	28	?	Chronic endometritis
*80*F	28	−27	72	56	45	30	254	Retroversion of uterus
Average		−22	60	40	31	15	510	

feature in common, namely, an abnormally low B.M.R., which varied from —13 per cent to —30 per cent. None of these individuals exhibited the usual clinical manifestations of thyroid insufficiency, and the results of the iodine tests did not conform with those obtained in clinically hypothyroid subjects. The highest 6-hour figure for the iodine tolerance test was 30 per cent of the injected iodine in the blood. Eleven of the 14 cases in this group yielded results within the accepted normal range of 9 to 23 per cent. Five responded after the manner of persons with hyperthyroidism.

The distribution of the percentages of the injected iodine remaining in the blood stream after 6 hours in the various groups of patients referred to above is represented in figure 4. Although there is a considerable spread as regards the control series, there is little overlapping of the values for this group with those for the thyrotoxic group of patients.

DISCUSSION

In determining these results, it is unknown at present what part, if any, is played by differences in the rate of the circulation of the blood. The speed of the blood flow is doubtless a factor to be considered, as mentioned by Perkin and Lahey (8). Some investigators (9, 10) have demonstrated the existence of an increase of the circulatory rate in exophthalmic goiter. Also, a decrease of the velocity of the blood flow has been found in myxoedema, but a normal rate of flow has been shown to exist in hypometabolism without myxoedema (11). To what extent the delayed disappearance of iodine from the blood stream in myxoedema is due to retardation of the circulatory rate and to what extent to the lack of fixation of this element by the thyroid gland is uncertain.

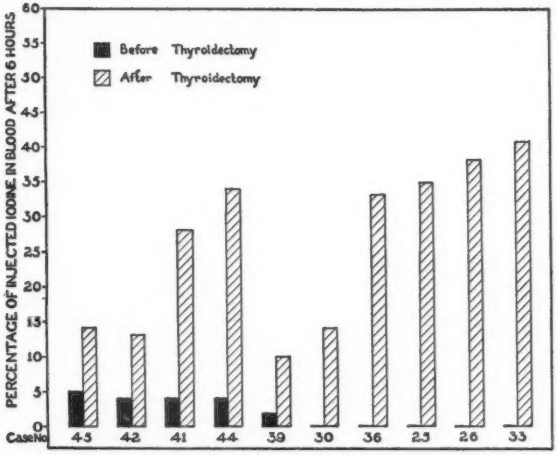

Fig. 3. Showing the effect of thyroidectomy on the results of the intravenous iodine tolerance test.

From the standpoint of diagnosis, however, this may not be a point of vital importance.

A noteworthy feature of the afore-mentioned observations is the absence of an absolute relationship between the B.M.R., and the results of the iodine tolerance test. Many factors, of course, may modify the basal metabolism. It is well known that the B.M.R. may be affected by influences apart from primary derangements of the functional activity of the thyroid and, in this regard, the anterior pituitary is doubtless of importance.

An interesting group of patients includes those classified as 'hypometabolism without clinical hypothyroidism.' It is observed in table 6 that 5 of these individuals showed an increased iodine tolerance, whereas the remainder gave values which might be interpreted as being normal. One may speculate indefinitely with respect to the paradoxical situation of a low B.M.R. accompanied by a rapid or normal rate of disappearance of injected iodine from the blood stream, but there may be some correlation between such clinical observations and the results of certain animal experiments.

Changes in the iodine metabolism corresponding to those noted in Graves's

disease in man were produced by Closs, Loeb and Mackay (12) in guinea pigs by injections of extracts of the anterior pituitary. Also, Friedgood (13) observed that the injection of anterior pituitary extracts into guinea pigs caused an initial rise of the basal metabolism followed by a remission in which the B.M.R. was low. It was suggested that prolonged stimulation may lead to impairment of the function of the thyroid gland with the development of hypothyroidism. Thompson and his co-workers (14) noted that the prolonged

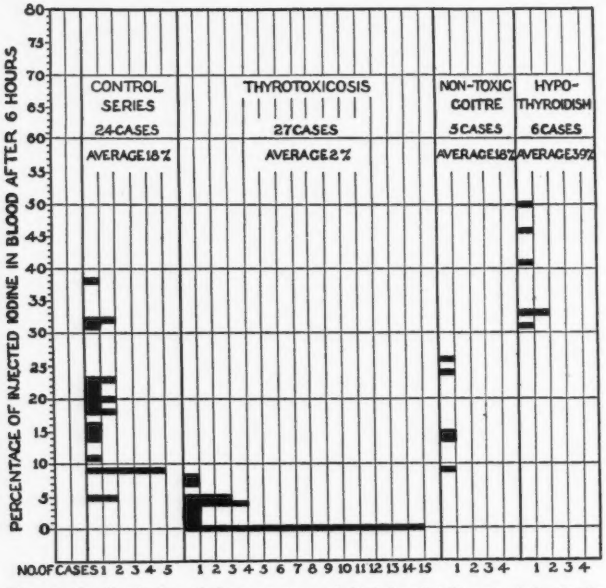

Fig. 4. Showing the distribution of the percentages of the injected iodine remaining in the blood stream after 6 hours in various groups of patients.

administration of APE in man caused only a temporary increase of the basal metabolism. The continuance of the treatment was accompanied by a return of the metabolic rate to the pre-treatment level and, in some cases, by a decline below that level.

There is, therefore, some justification for the belief that a type of thyroid dysfunction caused by imbalance of the pituitary control may be associated, simultaneously, with an increased or normal iodine tolerance and a low B.M.R. In other words, it is possible for a combination of structural hyperplasia to co-exist with functional insufficiency of the thyroid.

As outlined, the method of investigating the iodine metabolism by means of the intravenous iodine tolerance test is too complicated for ordinary clinical use, but it does introduce a different approach to the problems of thyroid disease.

SUMMARY

Anatomical and functional abnormalities of the thyroid gland may be considered from the standpoint of probable alterations of the iodine metabolism.

The nature of such disturbances can be studied by observing the rate and completeness of disappearance from the blood stream of iodine following its intravenous injection. The results of 91 iodine tolerance tests in 80 patients are recorded. The thyroid apparently is the only organ which abstracts and stores iodine from the blood. This property is accelerated in conditions of thyroid hyperplasia associated with thyrotoxicosis, as exemplified by the abnormal rapidity with which the iodine leaves the blood. Injected iodine disappears less rapidly from the circulation in hypothyroidism. The possible influence of variations of the circulatory rate in determining these effects is discussed. There is no definite correlation between the iodine tolerance and the B.M.R. The anterior pituitary through its influence upon the thyroid may affect the basal metabolism and the iodine tolerance test in an inverse manner. In conjunction with existing methods of clinical investigation the iodine tolerance test may permit a more comprehensive classification of endocrine disorders than heretofore.

REFERENCES

1. Watson, E. M.: Endocrinology **20**:358. 1936.
2. Elmer, A. W.: Endocrinology, **18**:487. 1934.
3. Marine, D., and H. O. Feiss: J. Pharmacol. & Exper. Therap. **7**:557. 1915.
4. Marine, D., and J. M. Rogoff: J. Pharmacol. & Exper. Therap. **8**:439. 1916.
5. Watson, E. M., and A. S. Barber: J. Lab. & Clin. Med. **22**:972. 1937.
6. Perkin, H. J.: Biochem. J. **27**:1078. 1933.
7. Von Bergmann, O.: Klin. Wchnschr. **6**:776. 1927.
8. Perkin, H. J., and F. H. Lahey: New England J. Med. **216**:501. 1937.
9. Fullerton, C. W., and G. A. Harrop, Jr.: Bull. Johns Hopkins Hosp. **46**:203. 1930.
10. Boothby, W. M., and E. H. Rynearson: Arch. Int. Med. **55**:547. 1935.
11. Macy, J. W., T. S. Claiborne and L. M. Hurxthal: J. Clin. Investigation **15**:37. 1936.
12. Closs, K., L. Loeb and E. M. MacKay: J. Biol. Chem. **96**:585. 1932.
13. Friedgood, H. B.: Bull. Johns Hopkins Hosp. **54**:48. 1934.
14. Thompson, W. O., S. G. Taylor, P. K. Thompson, S. B. Nadler and L. F. N. Dickie: Endocrinology **20**:55. 1936.

THE INFLUENCE OF THE THYROID GLAND AND OF THE OVARY ON THE METABOLISM OF IODINE

AN EXPERIMENTAL STUDY IN THE DOG

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In the course of a series of investigations concerning the level of iodine in the blood of dogs receiving Lugol's solution, certain experimental data were considered to indicate the existence of a correlation between the thyroid gland, the metabolism of iodine and the ovary. The experimental observations are detailed in the present report.

METHODS AND RESULTS

For the purpose of presentation, the experiments cited have been divided into 4 groups. *A.* The *influence of the daily ingestion of a specific amount of iodine on the blood iodine level of normal dogs.* Six female and 4 male dogs of the same breed and proportionate body weights (40 to 50 lb.) were given daily doses of Lugol's solution for periods ranging from 1 to 12 months. Lugol's solution, equivalent to 72 mg. of iodine, was administered by stomach tube daily at the same time. Prior to the giving of the iodine, 1 cc. of blood was withdrawn from the femoral vein for iodine analysis (1, 2). The animals received one meal a day, consisting exclusively of meat, immediately after the blood had been collected.

The results (fig. 1 and 2) showed that the blood iodine level of normal male and female dogs receiving daily 72 mg. of iodine represented graphically a series of peaks and dips varying from 750 to 2500 γ of iodine per 100 cc. of whole blood. The rise and fall of the level of iodine in the blood appeared to be more regular in female (fig. 1) than in male dogs (fig. 2). In one female dog which received iodine for a year, a very high peak of elevation in blood iodine occurred in the spring and again during the autumn. This peak may be attributed to seasonal influence, the period of fertility, or unknown factors (fig. 1).

Coincidental to the above, a normal animal became pregnant. The pregnancy followed a natural course with the animal continued on experiment. In this animal, the blood iodine level was appreciably depressed throughout the period of gestation (fig. 3). After the delivery of two normal pups, the blood iodine level rose, and maintained the same graphical relationship as previous to the pregnancy (fig. 3).

B. The influence of total extirpation of the thyroid gland on the blood

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iodine level of male and of female dogs ingesting the same specific amount of iodine daily. Four female and two male dogs from group *A* were subjected to complete thyroidectomy. Although the parathyroid glands were transplanted, the animals showed signs of hypoparathyroidism up to two weeks following operation. The attacks of tetany were treated by giving parathormone, together with calcium chloride and viosterol in milk. After 3 or 4 episodes were relieved, no further treatment was necessary. On complete recovery

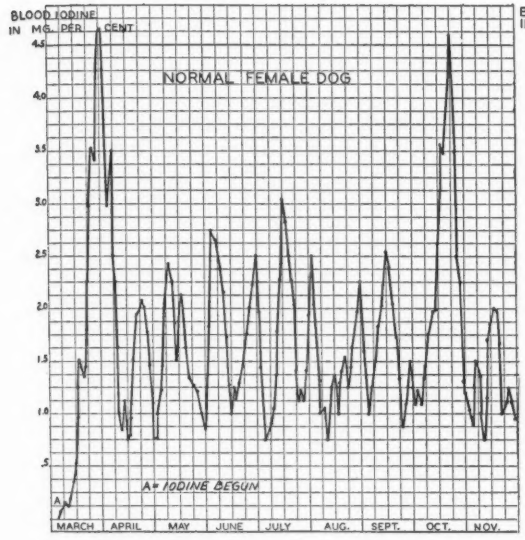

Fig. 1.

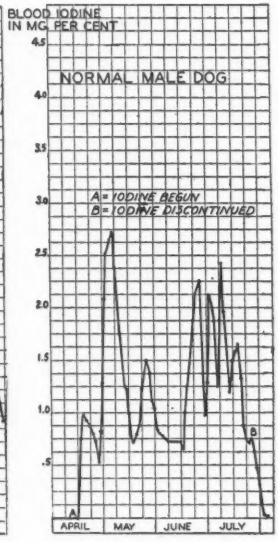

Fig. 2.

from the operation, the dogs were again given the same dose of iodine (72 mg.) daily, and their blood iodine levels were followed.

The results in female dogs revealed that the variations in the blood iodine level were unaffected by the removal of the thyroid gland (compare fig. 1 and 4). In contrast, the blood iodine level of male dogs showed a considerable decrease from that found prior to thyroidectomy (compare fig. 2 and 5), or in thyroidectomized female dogs (compare fig. 4 and 5). The comparative relationship as shown in figures 4 and 5 is believed to indicate the existence of a variation attributable to the sex of the animal.

C. The influence of extirpation of the ovary on the blood iodine level of thyroidectomized female dogs receiving 72 mg. of iodine daily. Bilateral oophorectomy was performed on two of the thyroidectomized female dogs. Following this procedure, iodine was again administered in the same daily dosage and the blood iodine levels were followed.

The results indicated that, following removal of the ovaries from thyroidectomized dogs, the blood iodine level fell in comparison with the previous level in the same thyroidectomized animal (compare fig. 4 and 6). The blood iodine graph of the female dogs on which oophorectomy and thyroidectomy

had been performed simulated that of the thyroidectomized male dogs (compare fig. 5 and 6). These findings indicated that the difference in the blood iodine level between male and female thyroidectomized dogs, noted in group *B*, was eliminated by the removal of the ovaries of the latter.

D. Iodine analysis of the tissues of dogs which had received iodine for varying lengths of time. At the conclusion of the above experiments, autopsy was performed on all of the animals, and iodine analyses were carried out on

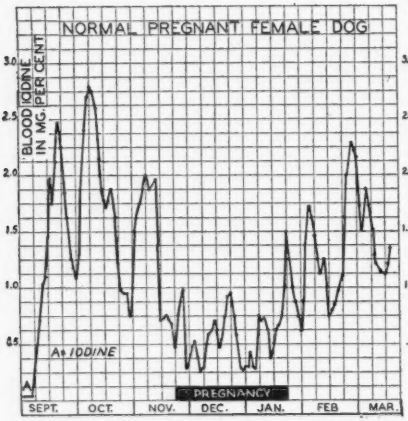

Fig. 3.

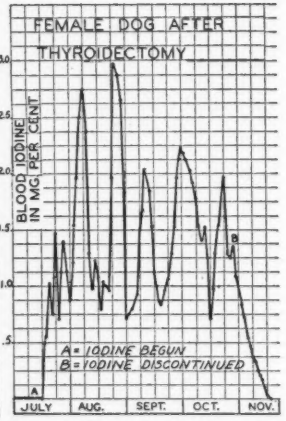

Fig. 4.

the various tissues. The values cited in table 1 were representative of the entire group.

From the table, it will be seen that the ovary contained approximately twice as much iodine as any other tissue, except the thyroid gland. Furthermore, the iodine content of the thyroid gland apparently increased with the period of time that iodine was administered.

DISCUSSION

In the present study, the more important factors affecting the level of iodine in the blood at any given time were considered to be *a)*, the amount of iodine administered, *b)*, the rate and extent of absorption, *c)*, the excretion and endogenous processes. In the first instance, it should be noted that the iodine administered daily was an excessively large amount to be given in a single dose. If divided doses had been given, a more cumulative effect than that obtained might have resulted. In view of the repeated doses and the large amount, the form of iodine used in these experiments was probably not important. Regarding absorption, it would seem logical to assume that the iodine administered was taken up quickly and completely into the blood, since unpublished data (3) showed that, following the administration of a single dose of iodine, the blood evidenced absorption within 5 minutes, with the feces devoid of a significant quantity of iodine until the third day. The complete absorption of the inorganic iodides of sodium and of potassium from the gastro-intestinal tract has also been noted by Greenbaum and Raiziss (4).

Under the present experimental conditions, it seemed reasonable to believe that the animal body would have become saturated with iodine, the excess being excreted. Since signs of iodism were not apparent in the animals, and analysis of the tissues revealed no marked accumulation of iodine, an equilibrium between absorption and excretion must have been established. Furthermore, the effect of absorption and excretion was minimized by taking blood for iodine analysis during the postabsorptive state. With these ideas in mind, the predominant influence on the blood iodine level appeared to be attributable to endogenous factors.

That the blood iodine level of normal dogs receiving 72 mg. of iodine daily fluctuated within a definite range seemed remarkable. This variation of rise and fall in the level was not due to experimental error, because an adequate number of iodine analyses indicated the trend of each variation in the level.

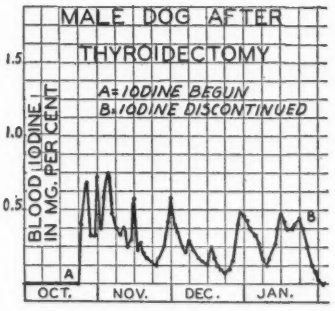

Fig. 5.

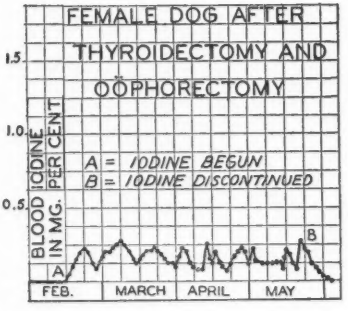

Fig. 6.

The results showed neither an accumulative effect nor a decreased tolerance to iodine. The fluctuations in blood iodine could be explained on the basis that the postabsorptive blood iodine level is dependent on the organic-inorganic iodine ratio in the blood. Such an explanation would necessarily assume that the thyroid gland, with its capacity to combine iodine in an organic form, would be the primary tissue affecting the control of the blood iodine level. The foregoing interpretation was not entirely supported by experimental data, since we have found that alcohol-insoluble fractions of the blood in the above instances gave iodine values of only one-sixth to one-twelfth of the total iodine content. However, the 'organic iodine fraction' did vary in quantitative relationship to the total blood iodine. With such an excess of alcohol-soluble iodine in the blood, it would appear that in these experiments tissues other than the thyroid gland must have exerted an effect on the blood iodine level. That tissues outside the thyroid gland affecting the metabolism of iodine may have been directly or indirectly influenced by the thyroid gland cannot be denied.

The comparative reduction of the blood iodine level in male dogs following thyroidectomy indicated that the thyroid gland is a primary factor in controlling the blood iodine. As pointed out above, the exact nature of this control is not clear. The presence of controlling factors outside the thyroid gland

was apparent because, under the conditions of the experiment, thyroidectomy in female dogs did not appreciably alter the blood iodine level. Since a comparative decrease was found in the blood iodine level, following oophorectomy in thyroidectomized female dogs, it follows that the ovary must have exerted an influence on the blood iodine level. In addition, the present findings indicated that the blood iodine level is influenced to the same degree by the ovary as by the thyroid gland, although this was not confirmed by removing the ovaries and leaving the thyroid gland intact. In this connection, the discovery

TABLE 1. THE IODINE CONTENT OF THE TISSUES OF DOGS RECEIVING IODINE DAILY.

	Mg. of iodine per cent (wet tissue)			
	A[1]	*B*[2]	*C*[3]	*D*[4]
Thyroid	18.3	27.6	31.2	34.6
Kidney	0.21	0.31	0.35	0.43
Liver	0.16	0.42	0.38	0.33
Lung	0.21	0.34	0.29	0.37
Heart muscle	0.24	0.33	0.32	0.30
Adrenal	0.19	0.52	0.41	0.29
Ovary	0.43	0.66		0.63
Testis			0.36	
Bile	0.20	0.28	0.27	0.26
Blood	0.24	0.36	0.42	0.46
Feces	0.20			
Intestine	0.18			

[1] *Animal A* received iodine daily for 1 month.
[2] *Animal B* received iodine daily for 4 months.
[3] *Animal C* received iodine daily for 7 months.
[4] *Animal D* reeceived iodine daily for 12 months.

of a proportionately high content of iodine in the ovarian tissue served as further evidence favoring an iodine-ovarian relationship, thus confirming the observations of Zoeppritz (5) and Weinberg (6).

SUMMARY

The blood iodine level of male and female dogs receiving 72 mg. of iodine daily was represented graphically as a series of peaks and dips varying from 75 to 2500 γ of iodine per cent of whole blood. The blood iodine level was considerably depressed following complete thyroidectomy in male dogs receiving iodine. In female dogs receiving iodine, complete thyroidectomy apparently had no influence on the blood iodine level. Following bilateral oophorectomy in the thyroidectomized female dogs, the blood iodine level decreased; the results simulated the values found in thyroidectomized male dogs.

Iodine analyses of the tissues of dogs receiving iodine for varying periods of time indicated that, exclusive of the thyroid gland, the ovary contained approximately twice the amount of iodine per gram weight of any other tissue.

REFERENCES

1. Perkin, H. J.: Biochem. J. **27**:1078. 1933.
2. Perkin, H. J., and R. B. Cattell: New York State J. Med. **36**:1033. 1936.
3. Perkin, H. J.: Unpublished data.
4. Greenbaum, F. R., and G. W. Raiziss: J. Pharmacol. & Exper. Therap. **30**:407. 1927.
5. Zoeppritz, B.: München med. Wchnschr. **2**:1898-1899 (Aug. 27). 1912.
6. Weinberg, B.: Beitr. z. Geburtsk. u. Gynäk. **19**:222. 1913-1914.

DYNAMIC TESTS IN THYROTOXICOSIS[1]

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There are two situations in the operative treatment of goiter in which the usual methods of estimating the patient require reenforcement, and it is significant that they represent two extremes of the disease. The highly thyrotoxic individual, whether he is the well-nourished patient continuously refractory to treatment, or the emaciated person who has lost over one-third of his average weight, always requires accurate estimation and is operated on in due time (frequently by a 2-stage procedure) as a questionable risk. A constant though small mortality rate obtains, especially when an accident adds to the calculated stress to which he has been subjected. On the other hand, we encounter many nervous, heart-conscious individuals with a goiter, some of them after extensive medication, and occasionally find it difficult to decide whether they are in a remission of recent thyrotoxicosis or whether their goiters have been involved in the course of a neurosis or of a pluriglandular syndrome. If the basal metabolism is normal in such cases, the objective evidence for accurate differentiation is scanty, and only prolonged observation may clarify the case. Thyroidectomy performed on an incorrect assumption of toxicity uniformly makes these patients more miserable than before.

METHODS AND PROCEDURE

In both of these situations we have been impressed with the value of dynamic tests, i.e., those which require work on the part of the patient, and in which he is not merely a passive spectator. The first of these is a determination of the ability of the patient to hold his breath under standard conditions and according to a definite technic (1, 2) to which we have adhered since 1928, although my father's original work with the test antedated that by several years. Breath-holding ability, by definition, is 'duration of voluntary apnea' (D.V.A.). Duration, in seconds, is determined after a maximal inspiration and, following a rest period, after maximal expiration. The result is then expressed as a fraction, of which the inspiratory phase is the numerator and the expiratory phase the denominator. The arithmetical value of this fraction, also of interest to us, is termed the index. The following example will help to clarify the derivation of these terms.

$$\text{D.V.A.,}\frac{\text{(inspiration) } 50''}{\text{(expiration) } 25''} = 2.0 \text{ index.}$$

[1] Read before the Section on Surgery, General and Abdominal, of the American Medical Association, Kansas City, Mo., May 14, 1936.

RESULTS

For purposes of investigation, these determinations have usually been made under basal conditions immediately following the estimation of basal metabolism, pulse rate and blood pressure. All observations, except those of basal metabolism, have been made by the author personally. Table 1 gives an outline of the changes occurring throughout the treatment of an average thyrotoxic patient, and of the expected improvement in the recorded criteria. Progressing ability to hold the breath roughly parallels improvement in basal

TABLE 1. *P.N.*, FEMALE, AGE 51.

Date	Pulse	B.P.	B.M.R.	D.V.A.	Index	Remarks
3/5	105	128/55–40	+34	33/21	1.6	Admitted
3/8	100	110/70		40/25	1.6	
3/16	84	120/80	+14	70/40	1.8	
3/17						Subtotal
3/26	90	110/80	−3	116/62	1.9	Discharged

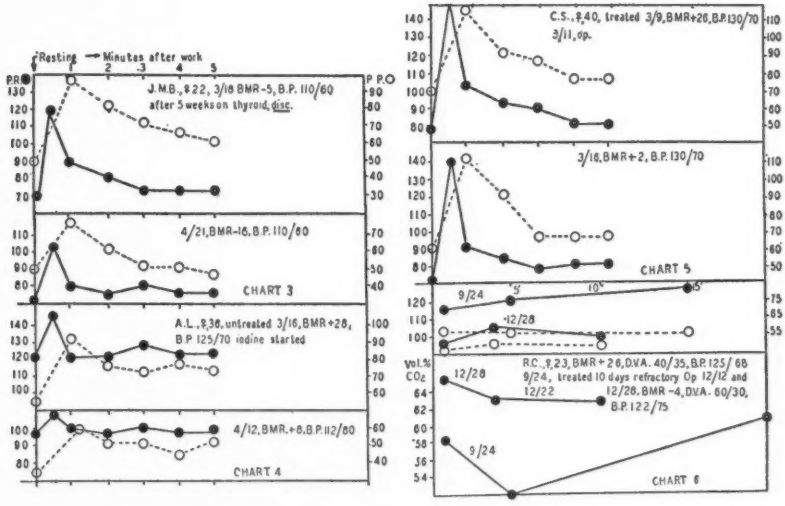
TABLE 2. SUMMARY OF OBSERVATIONS.

			D.V.A.	Index
			Seconds	
Group 1	For ligation	Admit.	27/19	1.4
		Pre-op.	33/23	1.4
2	For 2-stage.			
	(2 hosp.)	Admit.	35/24	1.5
	(1 hosp.)	Admit.	46/30	1.5
	1st stage	Pre-op.	47/29	1.7
	2nd stage	Pre-op.	57/34	1.7
3	For subtotal.			
	toxic	Admit.	41/31	1.3
	toxic	Pre-op.	62/36	1.7
	nontoxic	Pre-op.	72/40	1.8

metabolism, pulse rate and pulse pressure, although the correlation is not invariable; that it is not constant should not be surprising, since the ultimate mechanism determining ability to hold the breath is uncertain but seems to be the acidity of the respiratory center. We are interested in the relationship between the two factors, inspiratory and expiratory, that go to make up D.V.A., since it became obvious very early in this investigation that this relationship, which we express as the index, undergoes a characteristic change during the progress of the disease. With the technic of the test employed, duration after inspiration is about twice as long as after expiration in normal individuals, making the normal index 1.8 to 2.0; in acute, especially untreated, thyrotoxicosis, the two phases tend to equalize in duration and the index drops toward unity. As will be seen later, an index near unity is characteristic of unstabilized thyrotoxicosis, regardless of the duration of the phases in seconds; a reversion toward an index in the normal range of 1.8 to 2.0 occurs with improvement, since the inspiratory phase increases in duration more than does the expiratory phase.

In table 2, 178 observations on 111 patients are summarized, with the patients divided arbitrarily into 3 groups, according to the operative proce-

dures for which they were suitable; this gives a fair rating as to the severity of their individual cases. Data gathered on admission and at pre-operative examination in each group show the improvement registered in regard to D.V.A. We no longer consider ligation of the superior poles to have therapeutic effect, and have not practiced it since 1931. The average interval between admission and ligation in the patients in group 1 is 18 days; their refractoriness to treatment is illustrated by the small increase in duration and persistently low index. In group 2 are included all patients having 2-stage operation, whether at 1 or 2 hospital stays; the difference in duration in the admission data of the 2 subgroups is measurable, although they have the same unstabilized index (1.5). Those having both stages done at the same hospitalization are nearly all under 50 years, have had considerable previous treatment, and are able to withstand the first stage within a few days of ad-



mission; the interval between stages is 11 days. Those patients discharged after the first stage are usually over 50 years, and return for the second stage after an average of 95 days. It is clear that we have waited to do the second stage until the patient becomes, measurably at least, as good as before the first stage. The figures for patients in group 3 require no elucidation. The average interval between admittance of thyrotoxic patients and subtotal thyroidectomy (for those not previously hospitalized) is 8 days.

Since 1934 we have carried out another series of observations which has focussed our attention more sharply on the burden under which the thyrotoxic patient labors. The response of pulse rate, pulse pressure and the carbon dioxide combining power of the plasma to measured exertion (20 hops on each foot in 30 seconds) has been followed at intervals through the recovery period. Pulse rate and pulse pressure show an immediate and moderate elevation in the normal individual, and a return to resting levels in 2 to 3 minutes: the

carbon dioxide combining power shows not more than 2 volumes per cent deviation, practically the margin of error inherent in the method.

The early effect of overfeeding a normal individual with thyroid substance is shown in chart 3. On March 18th there were subjective manifestations of toxicity and the medication was stopped; on 4/21 the patient was once more comfortable. It is significant that the exaggerated immediate increase and delayed return to resting values on 3/18 were no longer present on 4/21, although the basal pulse rates and pulse pressures were identical. Chart 4 illustrates the response to treatment pre-operatively; although the pulse rate returned promptly to resting value on 4/2, pulse pressure had not returned after 5 minutes. Chart 5 shows data on a more severely thyrotoxic individual, who was, on 3/9, sufficiently stabilized for operation. Although the expected improvement in all details had occurred on 3/16, the fifth postoperative day,

TABLE 3.

Time	P.	B.P.	T.	R.	Bath
5:00 p.m.	180	60/ 0	105.6	28	98 F.
5:45	150	58/ 0	101.6	22	90
6:20	transfusion 650 cc.				
6:30	140	90/40	100.4	20	95
8:05	acacia 425 cc.				
8:30	135	115/50		20	94
9:05	135	125/40	99.4	20	94
10:30	140	80/40	99.6	20	96
12:00	135	110/40	99.0	20	96
12:30 a.m.	phonating; transfusion 500 cc.				
1:30	128	130/50	100.0	20	96
3:30	136	138/55	cyanotic		
5:00	100	80/20	100.0	20	98

it was obvious from the immediate over-response of the circulation to exertion that the patient should have continued limitation of all exertion. In chart 6 the early determinations were omitted for the sake of clarity; the largest deviations, therefore, do not appear. After prolonged treatment, 2-stage thyroidectomy was performed. The prolonged depression of the carbon dioxide combining power is obvious in the lower half of the chart.

Coller and Maddock (3) have shown that the fever of thyroid crisis is not associated with retention of water, but that the dissipation of body heat lags behind its production. If thyroid crisis were merely due to failure of the heat-dissipating systems, it should be possible to control such a reaction by aiding the faltering mechanisms. This we attempted by the use of the continuous neutral bath (90-94°F.), water being 27 times as efficient a conductor of heat as air. Table 3 gives excerpts from the protocol of one such patient, from the time of immersion in the bath, 6 hours postoperatively, until shortly before death. It is seen that the temperature of the patient can be brought readily to as nearly normal as is desired, and maintained there indefinitely. In our experience, with two spontaneous and one postoperative thyroid crises, since 1934, these patients continue on this fatal course and die with a normal temperature, although their needs for water, glucose and oxygen have been met. Some agent other than fever evidently has irreparably injured the vaso-

motor apparatus of such a patient, and a view that considers thyrotoxicosis as a disease merely of hypercombustion cannot be reconciled with these or with other facts.

SUMMARY

At certain points in thyrotoxicosis, our available 'static' methods of examination give us only incomplete information in regard to diagnosis and treatment. The author's test of breath-taking ability is presented with illustrations, also a tabulation and a graphic chart of its characteristic behavior in 178 observations of 111 goiter patients throughout the range of thyrotoxicosis.

D.V.A. (duration of voluntary apnea), determined in both inspiratory and expiratory phases, increases with improvement, as judged by other criteria; the relationship between the two phases (index) changes characteristically toward the normal, in which the inspiratory phase is about twice as great as the expiratory. The responses of pulse rate, pulse pressure and carbon dioxide combining power of the plasma after standardized exertion are discussed, four charts indicating the trends shown by 113 tests on 71 patients. These comprise an exaggerated immediate rise in pulse rate and pulse pressure and a profound fall in carbon dioxide combining power, with a slow return to resting levels. Less pronounced effects are seen in milder stages of the disease, but a persistence of the effects, especially in the pulse pressure, is usually found after basal metabolism, pulse rate and pulse pressure have returned to normal on treatment, with or without operation. As additional evidence that thyrotoxicosis is not purely a disease of hypercombustion, an illustrative protocol is presented of rapid reduction of body temperature, but ultimate death, by a continuous neutral bath in the case of a patient in postoperative thyroid storm. Similar therapy in less advanced cases is advocated.

REFERENCES

1. Bartlett, W., Jr.: Tr. Am. A. Study Goiter, P. 129, 1930.
2. Bartlett, W., Jr.: Tr. West. S. A., P. 550, 1935. Surg. Gynec. & Obst. **63**:576. 1936.
3. Coller, F. A., and W. G. Maddock: Tr. Am. A. Study Goiter, P. 188, 1933.

METABOLIC, CARDIOVASCULAR, AND BIOCHEMICAL CHANGES ASSOCIATED WITH EXPERIMENTALLY INDUCED HYPERTHYROIDISM IN SCHIZOPHRENIA

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From the wealth of data on the physiological processes in schizophrenia there has gradually crystallized in the minds of some investigators the inference that a disturbance of oxidation may characterize the disease process. In a recent paper bearing on this point, Hoskins (1) has presented a summary of the investigations carried out in schizophrenics at the Worcester State Hospital over a period of 10 years. In the evidence cited, reference is made to the incidence of thyroid dysfunction as one feature indicative of defective oxidation. The fact that there is a generally diminished metabolic rate and an apparently high tolerance to thyroid medication is adduced in this connection.

As a closely related problem, the present investigation has been concerned with the physiological responses of schizophrenic patients to prolonged hyperthyroidization. The nature of concomitant psychiatric changes in this group of patients will be reported separately. Although 'control experiments' which would indicate the comparative effects on normal individuals or patients with other types of psychoses are lacking, it has nevertheless seemed worth while to place these data on record because *A)*, as far as the writers are aware, this investigation is the first in which a systematic and prolonged study of the effects of administration of thyroid extract in extremely large amounts to human beings has been made, and *B)*, they offer some leads for the investigation of the schizophrenic disease process from the standpoint of metabolic activity. It is hoped, too, that the data may prove to be of value to those interested in the problem to what extent clinical 'hyperthyroidism' may actually be equated with true hyperthyroidism.

Table 1[1] presents a list of the physiological functions studied, the methods employed in the measurement of each, and the frequency with which observations were made. Desiccated thyroid (Armour's, U.S.P.) ingested orally was used throughout the investigation. In all cases it was administered under careful supervision.

Eight male schizophrenic patients, were selected as subjects because of advanced personality deterioration. These patients ranged in age from 24 to 37 years, and have been in the Worcester State Hospital from 4 to 20 years.

[1] The writers are indebted to Dr. J. M. Looney of the Biochemical Department and Mr. E. M. Jellinek of the Biometrics Department, and their staffs, for assistance in the collection and evaluation of the data.

The weights of the subjects at the beginning of the experiment ranged from 77 to 109 per cent of predictions from age and height (nutritional index).

The present investigation was begun on Oct. 27, 1935. It was carried until Oct. 13, 1936 (353 days) in 2 cases; until November 5, 1936 (376 days) in 5 cases; and until May 20, 1937 (572 days) in 1 case, an average period of 395 days. The statistical analyses refer all patients to the 353-day total period.

The physiological and psychiatric[2] observations were conducted on the same schedule throughout the investigation. This was divided into 5 periods: *a)*, the pre-medication period, lasting about 21 days; *b)*, the first medication period, during which the patients were started on 6 gr. desiccated thyroid (Armour's, U.S.P.) per day, and increased slowly by 3-gr. rises to a maximum

TABLE 1.

Physiological Function	Method of Measurement	Frequency of Observations
1. Basal Metabolic Rate	Benedict-Roth	Weekly
2. Body Weight	Clinical	Daily
3. Oral and Rectal Temperature	Clinical	Daily
4. Pulse Rate	Clinical	Daily
5. Blood Pressure (Systolic, Diastolic, Pulse Pressure)	Clinical	Daily
6. Circulation Time	Robb-Weiss	Weekly
7. Blood Morphology	Clinical	Weekly
8. Blood Sugar	Folin-Wu	Weekly
9. Blood Cholesterol	Myers-Wardell	Weekly
10. Total Nitrogen of Blood	Folin	Weekly
11. Total Nitrogen of Urine	Folin	Weekly
12. Urinary Creatine and Creatinine	Folin	Weekly

of 15 or 18 gr. per day, this period lasting roughly 95 days; *c)*, the inter-medication period, during which no thyroid was administered, this period lasting approximately 67 days; *d)*, the second medication period, during which thyroid was again given, beginning with 6 gr. per day and increased 3 gr. per day, every other day, until the maximum of the first medication period of 15 or 18 gr. per day was reached (which took from 9 to 12 days); this period lasted for approximately 69 days. In 1 patient, the thyroid dosage was increased by 3-gr. rises until a maximum of 36 gr. per day was reached. The medication period for him lasted 241 days; *e*), the post-medication period, during which no thyroid was administered, this period lasting about 101 days. At no time during this entire investigation was any other medication administered.

The patients were kept under strict supervision on the ward, and in fine weather were allowed 1 or 2 hr. daily outdoor exercise. Attempt was made to maintain the patients' weights, since it was felt that marked weight loss would militate against determining the specific significance of any changes. For this reason they were given extra feedings. Generally, thyroid medication

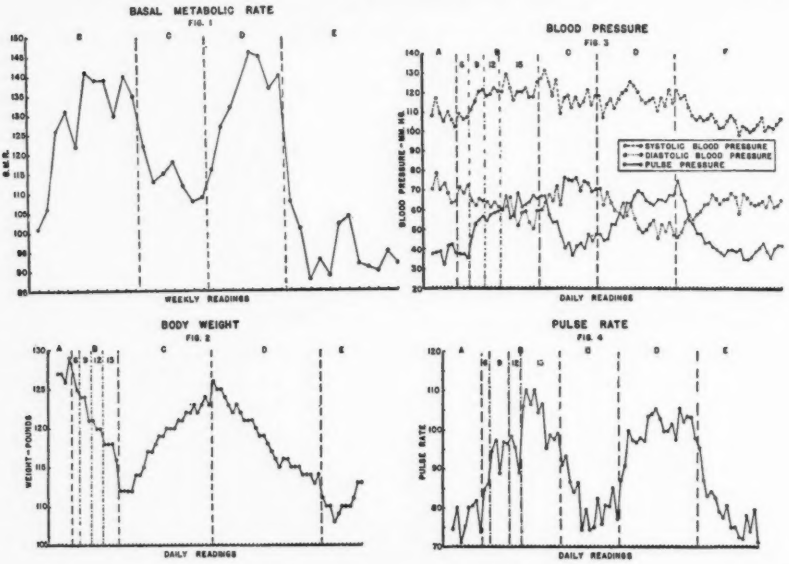
[2] The most striking psychiatric changes were in the sphere of motor activity. Marked speeding up of all activity was noted in all patients and torpor in some of them, sometimes to the degree of stupor; no changes in mood, in the so-called intellectual functions, or in other features of the psychotic picture were observed. It was felt that such mental changes were relatively minimal effects for the probable changes induced in cerebral activity (2), and as shown in thyroxinized patients in the electro-encephalogram (3).

was suspended when the patient's weight loss, despite forced feedings, reached 10 to 15 per cent of the initial body weight.

The literature bearing upon the specific variables investigated in relation to schizophrenia and the hyperthyroid states is presented in the separate discussion of the observations made on each of them. Due to limitations of space, only cursory accounts of the relevant literature can be presented.

RESULTS

The data were analyzed statistically, using the technic of the analysis of variance. Differences in the average levels of the various functions obtaining



among the periods described above, as well as among individuals, were tested for significance. Likewise the trends were tested for differences by periods and individuals. The term 'significant difference' will be used throughout in its statistical connotation. Differences, whether in average levels or trends, are designated as 'significant' when the probability of their occurrence through chance is not greater than 5 in 100 trials.[3]

1. Basal metabolic rate. Variation of the oxygen consumption rate (designated B.M.R.) might readily be expected to occur with thyroidization. Our data, presented graphically in figure 1, indicate that after one week of medication the average B.M.R. was increased from the pre-medication value of 95 to 101, and after 2 weeks to 106 per cent of prediction. The third week witnessed a rise to the average of 126. The maximum of 141 was reached after 6 weeks. After this, for the next 7 weeks, except for random fluctuations, the

[3] Readers interested in the primary data and their statistical analysis may obtain mimeographed copies on request from the authors.

curve remained at about the same level. It is of interest that the maintenance of the curve at a given high level persists for such a long period, despite the expected cumulative effect of thyroid. In the inter-medication period the curve dropped, but not very abruptly. This was of the order of 12½ points. Although the inter-medication period lasted 9 weeks, this was apparently not long enough for the initial B.M.R. level to be restored. The lowest point reached during this period was 108. Thus the second medication period started from a higher level than the first medication period. The maximum B.M.R. of 146 of this period, characterized by rapid increase of dosage, was 5 points higher than that of the first medication period, and was reached in 5 weeks. This maximum is an increase of 38 points, compared with the base of the second medication period, whereas in the first medication period an increase of 46 points compared with its own base occurred. Thus one cannot say that the rapid increase of dosage brings about a sharper rise and an earlier reached maximum. Nevertheless, the trends of the weekly means, displayed in the 2 medication periods, show a statistically significant difference. This is mainly due to the flattening of the trend in the first medication period after the sixth week, whereas in the second medication period there was a falling off of the effect after the sixth week.

Even greater than the differences between the first and second medication periods were the differences between the inter-medication and the post-medication periods. After the second medication period the falling off of the B.M.R. was much sharper than after the first medication period, a drop of 32 points in the first week as compared with a drop of 12½ points in the first week after the first medication period. It is difficult to say whether this difference is due to a desensitization to thyroid, or whether it must be attributed to the fact that the first medication period lasted 13 weeks, while the second medication period lasted only 9 weeks. At the end of the third week of the post-medication period, the B.M.R. dropped even below the average level of the pre-medication period, but this 'overthrow' is not significant.

2. *Body weight.* Loss of weight as a feature of hyperthyroidism is one of the most characteristic signs. One might expect that in the present study the increased metabolic rate would be reflected in weight loss, and as already noted, to guard against this, extra feedings were part of the experimental regimen. In all patients, a ravenous appetite developed during the first medication period and lasted well into the inter-medication period. Appetite was again increased during the second medication period, and persisted into the post-medication period.

The weight changes for the group are presented graphically in figure 2. The weight loss and gain during the medication and non-medication periods respectively are obvious. It should be pointed out, however, that the graph of the post-medication period is not representative, since the heaviest patient was not included in these data because medication in his case, as noted above, was continued throughout this period.

3. *Body temperature.* The data of daily oral and rectal temperature readings (for brevity sake not presented) manifested merely random variation in

the medication and non-medication periods. It would therefore appear that under these experimental conditions the effects of hyperthyroidization on body temperature are essentially nil. This finding tallies largely with the observations of Joll (4).

4. Blood pressure. Although the literature dealing with the question of blood pressure variations in disturbances of the thyroid gland is not concordant, it appears that generally thyrotoxicosis is associated with an increased systolic pressure and an increased pulse pressure (5, 6, 7).

a), Systolic pressure. The blood-pressure data of this investigation are presented graphically in figure 3, in which rather large increases in systolic pressure may be observed. The average systolic level before medication was 109 mm.Hg, and in the first 3 weeks with daily dosages of 6 gr. did not produce any significant rising trend. However, in the following 4 weeks with daily dosages of 9 gr., there was a significant rising trend, bringing the blood pressure to a peak, the average of which is 122 mm.Hg. After this the curve flattened out, and there was no further significant trend, even when the dosage was increased to 12 gr. per day. Under 15 gr., however, another rise occurred, bringing the maximum to 130 mm.Hg. However, this maximum was not significantly different, and the level dropped back immediately. One cannot therefore, speak of a significant blood pressure trend during the administration of 12 or 15 gr. per day. Individual patients differed significantly from each other in their average levels within each dosage group, but they differed from each other most in their averages under 15 gr.

In the inter-medication period, the systolic pressure dropped slowly. The declining trend was significant. However, it did not re-attain the pre-medication level except in one instance; towards the end of the inter-medication period the blood pressure was about 6 points higher than in the pre-medication period. Thus here, as was the case with the B.M.R., the second medication period started with an initially higher systolic level. In this period the systolic pressure rose much faster than in the first medication period, and reached a maximum of 126 mm.Hg., after which it fell off again rapidly; in the last days of the second medication period the average was about 116 mm.Hg. The trend between the first and second medication period was significantly different. As was the case with the B.M.R., the greater and earlier maximum observed in the second medication period could probably be attributed to the fact that the base-line of this period was higher than in the first medication period. In the post-medication period the falling off of the systolic pressure was rapid, and went below the level of the pre-medication period. The trends of the inter-medication and post-medication periods were significantly different.

b), Diastolic pressure. The levels of the diastolic blood pressure moved opposite to those of the systolic, but here the trend became significant only under the daily dosage of 15 gr., whereas for the systolic pressure a significant trend was found under 9 gr. For the total range of medication the declining trend of the diastolic pressure was significant. Also here the difference between the average levels of individuals increased as the medication went on. In fact, the differential individual response to dosages was even more pro-

nounced in the case of the diastolic pressure than in that of the systolic pressure. The average effect of the second medication period on the drop of the diastolic pressure was significantly greater than that of the first medication period, and the trends differed from each other significantly. Whereas the reaction of the systolic pressure in the second medication period was diphasic, the reaction of the diastolic pressure was constant during this whole period.

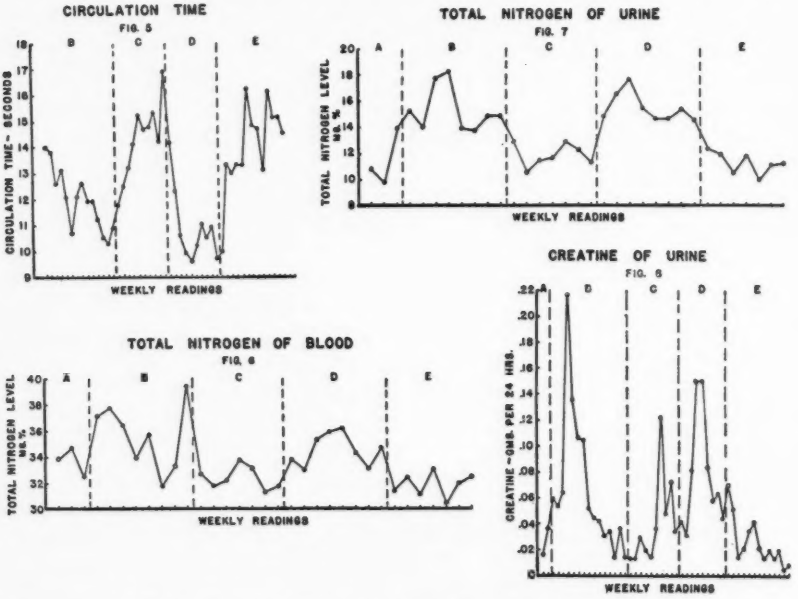
c), Pulse pressure. In view of the findings in the variations of the systolic and diastolic pressure, it is clear that the pulse pressure levels would manifest significant trends in both the medication and non-medication periods. As to dosage groupings, the trend began to grow significant under 9 gr. per day, and flattened out more or less under 12 or 15 gr. These responses were significantly different from individual to individual, and as in the case of systolic and diastolic pressure the pulse pressure, too, differed from individual to individual most under dosages of 15 gr. per day. The differences between the inter-medication and post-medication periods were the same as those found in the case of the diastolic pressure.

5. *Pulse rate.* That there is a parallelism, if only a moderate one, between the degree of tachycardia and increase of B.M.R. in hyperthyroidism has been shown by such investigators as White and Aub (8), Blumgart, Gargill, and Gilligan (9), and Means and Aub (10). Variation in the pulse rate differed in some respects from that of the blood pressure; the blood pressure responses throughout the experiment were different from individual to individual, and manifested increasing heterogeneity with increasing medication. The pulse rate, on the other hand, became more homogeneous with increasing medication. Furthermore, it may be observed from figure 4 that the pulse rate trend did not flatten out under 15 gr., but fell off while medication was still proceeding. In the inter-medication period, the initial level of 75 was reached towards the end. Thus, unlike those of B.M.R. and blood pressure, the level of pulse rate at the beginning of the second medication period was the same as that of the first medication period. In the second medication period the maximum was lower than that reached in the first medication period, namely, 105 as against 110. Although this maximum was reached a few days earlier, the difference of time was not significant. The rising trends of the first and second medication periods were significantly different from each other, but the falling trends of the inter-medication and post-medication periods not so.

6. *Circulation time.* Studies of circulation time have been reported by various investigators, most recently by Macy, Claiborne, and Hurxthal (11), who maintain from their review of the literature and their own data that the velocity of blood flow is generally increased in hyperthyroidism. The data of the present investigation are presented graphically in figure 5. The circulation time was not studied during the pre-medication period. In both medication periods, as may be seen, a decrease in circulation time occurred, and in the inter-medication and post-medication periods an increase. The inter-individual variation was increased during the non-medication periods, indicating a persistence of the after-effects of thyroid on the circulation time. Whether or not the trend in the second medication period is essentially different from that of

first medication period cannot be said, since there was no base-line to which the values obtained in the first medication period may be referred. On the whole, one may conclude from these data that the effect of hyperthyroidization is to speed up the circulation time considerably, and that in some individuals there is a rather prolonged after-effect.

7. *Blood morphology.* The most relevant paper on the effects of ingested thyroid on the blood morphology in man (schizophrenic patients) is that by



Hoskins and Jellinek (12). They found that, following thyroid medication, the average erythrocyte counts of the entire group were increased significantly, whereas the leucocyte counts decreased slightly. In the case of 52 patients initially anemic, the erythrocyte counts at first increased, then decreased as the dosage was raised; a similar diphasic effect on the leucocytes was also noted.

From data of the present study (not presented) it was seen that the erythrocyte count generally underwent the same diphasic reaction to thyroid substance, as has been reported by Hoskins and Jellinek (12). The data of the present study, however, were much less definite, probably owing to the smallness of the sample. Random fluctuation of the erythrocyte count during the first medication period was marked, and it was only towards the end of this period that a strong rise was noted. The erythrocyte count rose from an average of about 4,900,000 to a maximum of 5,200,000. During the inter-medication period the level dropped, but not to the general level of the pre-medication period. During the second medication period the level dropped sharply to that of the pre-medication period. This tallies with an observation made by Hoskins and Jellinek (12) that if the erythrocyte count is above 5,000,000

when thyroid medication is begun the count becomes decreased under medication.

The leucocyte count manifested too much random variation for a sample of this size to permit of any conclusions.

8. Blood sugar. Mirsky and Broh-Kahn (13), in a recent paper remark. "Although it is acknowledged by many that hyperthyroidism is associated with a profound disturbance in carbohydrate metabolism, the nature of the derangement is obscure." They state as well established (14) that in animals thyroid feeding is followed by depletion of glycogen stores. In their own experiments they observed that in thyroidized, eviscerated animals, the blood-sugar level was lowered more, and the rate of disappearance of blood glucose was swifter than in non-thyroidized animals.

Only the fasting blood-sugar level was studied here. The data (not presented) indicated that much random variation of blood-sugar level occurred. Furthermore, there was a suggestion of a general downward trend throughout the investigation, irrespective of the presence or absence of medication. But because of the great variation, this could not be statistically validated.

9. Blood cholesterol. Looney and Childs (15) have reviewed the literature of the cholesterol levels in schizophrenia, and find that there is a general lack of agreement. They attribute the diversity of observations to the great variation in the same individual, even from hour to hour, as well as to variation in the methods employed. From their own extensive study, lasting over 3 separate periods of 1 month each, they have concluded that schizophrenic patients tended to have a lower blood cholesterol content than normal individuals. They observed no relationship between the cholesterol level and the B.M.R. On the other hand, Hurxthal (16) observed in a series of 505 patients with thyroid disease that the lowest average cholesterol values occurred in patients in or near thyroid crises, in exophthalmic goiter, and recurrent hyperthyroidism. On the average, the blood cholesterol level and the B.M.R. were in 'reciprocal' relationship to each other.

In the present study (data not presented) it was found that even under conditions of extreme hyperthyroidization no marked displacement of the seasonal trends occurred. Apparently the effect of thyroid substance on the cholesterol level is not sufficiently strong to disturb the seasonal equilibrium, even in the presence of an increased oxidative rate.

10. Total nitrogen of the blood and urine. Although Meyers (17) makes the statement that thyroid illness produces no appreciable alteration of urinary nitrogen excretion, Bodansky and Duff (18) have demonstrated experimentally that the administration of thyroxin or desiccated thyroid to rats was followed by increased nitrogen excretion. It would be expected that, thanks to the increase of metabolism due to thyroid, the total protein level would be raised. The data of the present study bear out such an assumption.

From figure 6 it may be seen that the average blood-nitrogen level of 33.7 during the pre-medication period rose significantly in the first medication period to about 38.0, then dropped, but reached a maximum of 39.4 towards the end of this period. During the inter-medication period, the level dropped

sharply to somewhat below that of the pre-medication period. In the second medication period, the average level rose again, more slowly than in the first medication period and not so high, coming to a maximum of 36.0. The rising trends of the first and second medication periods were significantly different from each other. In the post-medication period the values again fell, and even went below the level of the pre-medication period.

Data pertaining to variations of the total nitrogen of the urine are presented graphically in figure 7. The trends were very similar to those of the blood nitrogen but more definite, since they were subject to less random variation. The values rose from an average of 11.5 in the pre-medication period to a maximum of 18.0 in the first medication period. During both non-medication periods the levels dropped back to the values of the pre-medication period. All trends were significant.

It appears likely, therefore, that the increased blood and urinary nitrogen reflected in part the increased food intake during the medication periods (exogenous metabolism), but the nitrogen level dropped at a time when the food intake remained high, thanks to the persistence of the voracious appetite well into the non-medication periods.

11. Urinary creatine and creatinine. Spontaneous creatinuria in thyrotoxicosis has been reported by Palmer (19) and Hunter (20), among others. The production of creatinuria by thyroid feeding as chronicled by earlier investigators has been confirmed by Carson (21). Bodansky and Duff (18) have demonstrated that in thyroxin-fed rats creatinuria is increased; only random fluctuations of the creatinine output were seen. Forschbach (22) found 'strikingly slight' amounts of urinary creatinine in patients with Graves's disease. It might well be assumed that the effects of thyroid feeding would be reflected as an increase in endogenous (creatine-creatinine) metabolic changes.

The data bearing on the variations in creatine level are presented in figure 8. It may be seen that during both medication periods the creatine values rose significantly, dropping back to the pre-medication levels in the non-medication periods. All the trends were highly significant, as were the differences between the trends. The creatinine levels (data not presented) showed only random variation.

The data indicate that under such conditions of experimental hyperthyroidism, an increase of endogenous (creatine) metabolism occurs.

DISCUSSION

These observations have been placed on record because of the uniqueness of the data, derived from the administration of unusually large doses of thyroid over prolonged periods of time to human subjects. The fact that these patients suffered from schizophrenia is of additional importance. In none of the patients (including one in whom medication was raised progressively to, and maintained at, a maximum of 36 gr. per day) were ill effects observed. How far the conditions of the present investigation, with respect to amount and duration of thyroidization, would be safe for non-schizophrenic individuals is problematic. It is, of course, well known that the mere feeding of thyroid

extract to human subjects is not sufficient for the experimental production of Graves's disease. The lack of production of any physical symptoms such as excessive perspiration, gastro-intestinal disturbance, tremor, pupillary changes, or exophthalmos is very striking. On the other hand, as demonstrated by the present data, quite striking changes in some of the metabolic, cardiovascular, and biochemical functions can be observed. It therefore seems plausible to argue on the basis of these findings that in schizophrenics there may be present factors of resistance, or barriers to the expression of increased oxidation, in the usual clinical features of hyperthyroidism.

SUMMARY

The physiological changes in 8 male schizophrenics subjected to prolonged periods of large doses of dessicated thyroid (Armour's, U.S.P.), ingested orally, are reported. The changes were compared with the observations on the same variables made before and after the medication periods, which consisted of slow and rapid thyroidization respectively to a maximum of 15 or 18 gr. per day. It was found in the patients as a group that the B.M.R. rose quite markedly in both medication periods, and dropped during the non-medication periods. Despite the induced ravenous appetite and extra feedings, the body weight dropped during the medication periods. Increased appetite persisted well into the post-medication periods.

Oral and rectal temperature readings showed only random variation throughout the whole period of investigation. The systolic blood pressure increased under medication, and dropped when medication was suspended; the systolic rise was greater, and reached an earlier maximum during the period of rapid medication. The diastolic pressure decreased during the medication period, a trend which was significant only under maximal dosage, but the general downward trend was also significant. The pulse pressure increased significantly under medication. Marked inter-individual differences in these variables were observed. The pulse rate manifested a rise under medication, the patients individually becoming more homogeneous with respect to this variable, as the dosage was increased. This latter observation was in contrast with that made for blood pressure, in which the patients' homogeneity decreased as dosage was increased. The circulation time underwent a decrease under medication, despite considerable random variation. The inter-individual variability was greatly increased following the second medication period.

Blood morphology studies revealed that under medication there was a slight rise of the erythrocyte count, which manifested a diphasic reaction. The leucocyte counts were impossible to evaluate because of the great variability. The blood-sugar level manifested considerable variability, and a general suggestion of a downward trend throughout the investigation could not be validated. The blood-cholesterol level underwent no changes except for seasonal variation, which thyroidization, apparently, could not displace. The total nitrogen level of the blood rose during the medication periods. The total nitrogen level of the urine also manifested a rise under medication, but in a more definite manner than the blood nitrogen level, because of less random

variation. The blood-creatine level manifested a rise in the medication periods. The creatinine level underwent no significant changes.

REFERENCES

1. Hoskins, R. G.: Arch. Neurol. & Psychiat. **38**:1261. 1937.
2. Santenoise, D.: Pneumogastrique Appareil Thyroïdien et Activité Cérébrale, Le Grand, Paris, 1929.
3. Rubin, M. A., L. H. Cohen and H. Hoagland: Endocrinology **21**:536. 1937.
4. Joll, C.: Diseases of the Thyroid Gland, Whitefriars Press, London, 1936.
5. DeCourcy, J. L.: J. Kansas M. Soc. **30**:44. 1929.
6. Boothby, W. M.: Cited by Davies, H. W., and J. Eason (7).
7. Davies, H. W., and J. Eason: Quart. J. Med. **18**:36. 1924.
8. White, P. D., and J. C. Aub: Arch. Int. Med. **22**:766. 1918.
9. Blumgart, H. L., S. L. Gargill and D. R. Gilligan: J. Clin. Investigation **9**:69. 1930.
10. Means, J. H., and J. C. Aub: Arch. Int. Med. **24**:645 1919.
11. Macy, J. W., T. S. Claiborne and L. M. Hurxthal: J. Clin. Investigation **15**:37. 1936.
12. Hoskins, R. G., and E. M. Jellinek: Endocrinology **16**:455. 1932.
13. Mirsky, I. A., and R. H. Broh-Kahn: Am. J. Physiol. **117**:6. 1936.
14. Coggeshall, H. C., and J. A. Greene: Am. J. Physiol. **105**:103. 1933.
15. Looney, J. M., and H. M. Childs: Arch. Neurol. & Psychiat. **30**:567. 1933.
16. Hurxthal, L. M.: Arch. Int. Med. **51**:22. 1933; *ibid.*, **52**:86. 1933; *ibid.*, **53**:762. 1934.
17. Meyers, M. K.: Cited by Falta, W., Endocrine Disease, Blakiston, Philadelphia, 1923.
18. Bodansky, M., and V. B. Duff: Endocrinology **20**:822. 1936.
19. Palmer, W. W.: Proc. Soc. Exper. Biol. & Med. **25**:229. 1927.
20. Hunter, A.: Creatine and Creatinine (Monographs on Biochemistry), Longmans, London, 1928.
21. Carson, D. A.: Proc. Soc. Exper. Biol. & Med. **25**:382. 1928.
22. Forschbach, J.: Cited by Falta, W., Endocrine Diseases, Blakiston, Philadelphia, 1923.

EXPERIMENTAL INDUCTION OF ESTRUS IN THE DOG[1]

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It has been demonstrated that adrenalectomized and immature female dogs may be brought into estrus with an extract of menopause urine (1, 2). The induced heat was of normal duration, and was apparently followed by a typical metestrum, as indicated by the prolonged survival period after cortical hormone had been withheld. These results compare favorably with those obtained when dogs are adrenalectomized during normal estrus (3), as well as when previously adrenalectomized dogs spontaneously come into estrus (4). Estrus has also been induced in the dog with follicular ovarian hormone (5 to 7). However, Grollman (7) found that adrenalectomized dogs following estrus induced with theelin failed to exhibit a prolonged life span, and he concluded that the induced cycle failed in some respect to duplicate all the physiological changes of normal estrus.

Previous experiments with an extract of menopause urine were performed with specific reference to the effect of the pseudopregnant period on the life span of adrenalectomized dogs, and therefore a study of this hormone's effect on the estrus cycle of the normal dog together with associated phases of mating, ovulation, and the general effect on the ovary and uterus seemed pertinent to investigation. Furthermore, it appeared of interest to compare the estrus induced with menopause urine extract and also estrone to the spontaneous estrus cycle.

MATERIAL AND METHODS

The experiments were performed on 20 female dogs, and in addition 5 mature animals were sacrificed for controls. Sixteen dogs were used for the experiments with an extract of menopause urine, 'gamone'.[2] Fourteen of these dogs were mature, either in the anestrus or metestrus phase, whereas 2 were immature, approximately 8 months of age. The extract was dissolved either in distilled water or 90 per cent glycerine, injected subcutaneously. The daily dosage was varied, but in later experiments a daily dosage of 75 R.U. proved satisfactory.

Estrone (progynon-B, in oil) was used on 4 mature dogs. Three normal dogs were in an anestrus state, whereas one dog was castrated with removal of the ovaries and a large part of the uterus. Injections were made subcutaneously in a daily dosage approximating 100 R.U. per 4 kg. body weight.

[1] Presented to the faculty of Princeton University in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

[2] Menopause urine extract 'gamone' and follutein were supplied by E. R. Squibb & Sons through the kindness of Dr. J. A. Morrell. Progynon-B was supplied through the courtesy of the Schering Corporation.

The control animals were in the laboratory a sufficient length of time to allow accurate determination of their phase in the estrus cycle.

Examination as to vulva swelling, hemorrhagic discharge and mating with normal males was carefully made. Using the blood smear technic daily vaginal smears were taken and stained with methylene blue. The ovaries, uterus and part of the vagina were removed from all dogs that were sacrificed.

The estrus cycle of the normal dog. The spontaneous estrus cycle of the dog has been studied by many investigators, but the most thorough experimental study is that of Evans and Cole (8). To allow adequate comparison with the induced cycle as to vaginal smears and the histology of the genital organs, 5 dogs were sacrificed at different phases of their normal cycle. The vaginal smear changes and the condition of the ovaries, uterus and vagina were found to be entirely in agreement with the previous study (8).

Experiments on the induction of estrus with menopause urine extract. That the urine of women past the menopause, of castrate women, as well as of individuals with certain types of genital tumors contains an APL gonadotropic factor was first demonstrated by Zondeck (9) and, since that time, has been confirmed by numerous investigators. Many have described pure follicle stimulation responses for an extract of menopause urine (10 to 15). On the other hand, luteinizing effects have also been shown to follow injections of this material (16 to 22). Leonard and Smith (11), using hypophysectomized rats, clearly demonstrated follicular stimulation, and this is supported by Tyndale and Levin (23), who find, however, that some luteinization resulted with the normal rat. Frank and Salmon (22) suggest that at least 3 factors influence the production of luteinization in the normal immature rat, namely, the method of extraction, the quantity of extract administered and the time. Induction of estrus in the dog, using this hormone, was primarily related to the adrenalectomized animal (1, 2).

The dogs in the first series of 'gamone' experiments were injected until they would first accept the male. In all 6 cases swelling of the vulva, bleeding (8 excluded) and mating was successfully induced. Vulva swelling always reached a maximal state. Pro-estrus bleeding, usually evident shortly after the first vulva swelling and often profuse, was generally of a somewhat shorter period than the normal. Using the criterion of mating as indicative of estrus, this period was found comparable to the normal, being 8 to 9 days in duration. Vaginal smears, taken daily through all phases of the induced cycle, were found to correspond with those of the spontaneous cycle.

Five of the first 6 dogs were sacrificed at various phases for histological comparison with the controls. The vagina was typical in all respects to the control animals in each phase. The uterine endometrium of the 2 dogs in estrus was hypertrophied, and exhibited a glandular condition of the normal estrus type. In early metestrus the endometrium was identical to the control (fig. 1, 2). The 3 zones, that is, the crypts around the lumen, the clear zone and the marked increase in glandular complexity in the basal region were quite evident. After 15 days in pseudopregnancy, the endometrium was still hypertrophied but showing signs of regression, and by the end of the period had regressed to an anestrus condition.

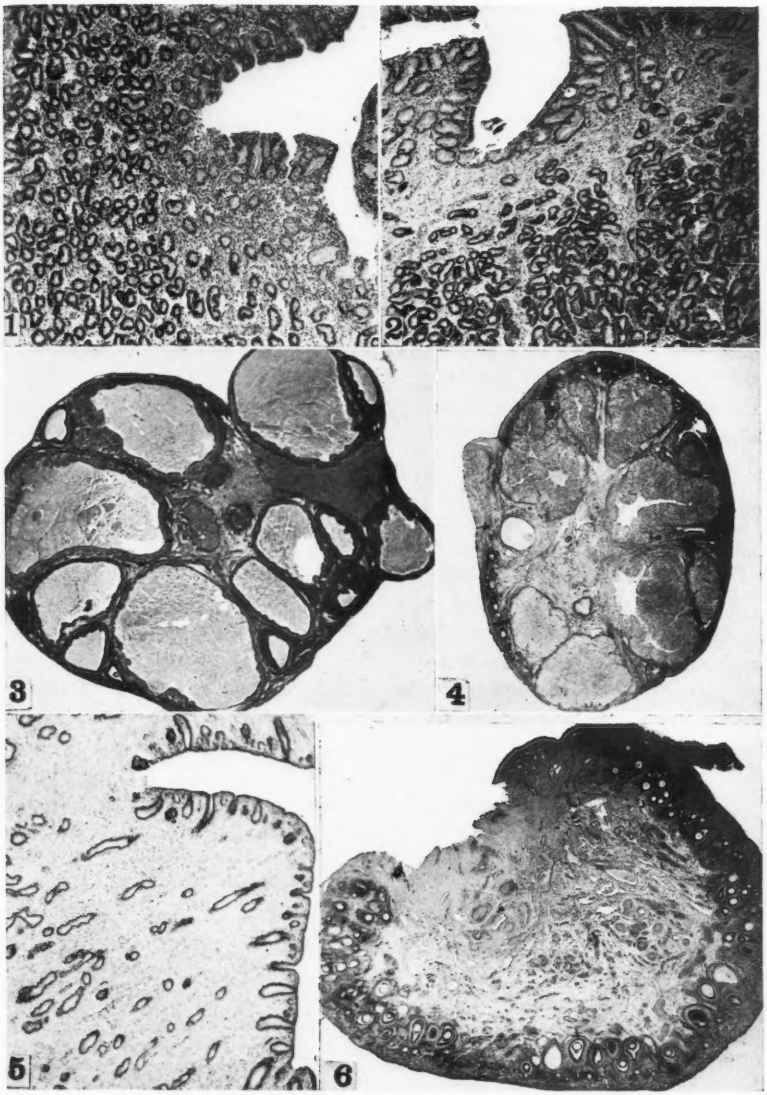

Fig. 1. Early pseudopregnant endometrium following estrus induced with menopause urine extract. x34.
Fig. 2. Early pseudopregnant endometrium following spontaneous estrus. x34.
Fig. 3. Ovary during early estrus induced with menopause urine extract. x3.
Fig. 4. Ovary 15 days in pseudopregnancy following estrus induced with menopause urine extract. x6.
Fig. 5. Endometrium 10 days after estrus induced with progynon-B. x34.
Fig. 6. Ovary 10 days after estrus induced with progynon-B. x9.

The ovaries of the dog in estrus were 'mulberry-like' on gross examination, with 65 follicles on the 2 ovaries, the average weight of these organs being 5.1 gm. in comparison to 1.3 gm. for a control in estrus. Several follicles were hemorrhagic, but ovulation points were absent. The ovaries revealed extensive follicle stimulation, although in some cases with slightly thickened granulosa, whereas in other follicles the wall was only 2 to 3 cells in thickness. Normal ova were found to be present in many cases, either as a discus poligerus or corona radiata. Three fair-sized compact corpora lutea were present, but this can readily be explained, since the animal was in early metestrus when the experiment was begun (fig. 3).

The ovaries of an immature dog (7) were not 'mulberry-like' but appreciably increased in size, averaging 2.1 gm. in weight. Previous investigations on normal immature rats indicated that high dosage frequently resulted in a luteinizing effect, and since this dog had received a fairly high dosage over a long period of time, any luteinizing action of this extract would be evident. Furthermore, the dog was immature, so that the complication of corpora remaining from a previous estrus was avoided. The ovaries exhibited pure follicle stimulation in exclusion of any thickening of the granulosa, but it is of interest, in this case, that the ovary never had more than 4 large follicles in a section, unlike the mature dog in which 8 or 10 were found. In fact, the center of the one ovary was composed entirely of 2 enormous follicles comprising the total area of the section. Normal ova were found to be present.

In order to observe the expected luteinizing action of the animal's own pituitary, another dog was sacrificed on the fifth day of pseudopregnancy. This dog had been in estrus 4 months previously, and was chosen for this experiment because residual corpora would be absent. The ovaries were found to contain large normal appearing corpora, sufficient to maintain a pseudopregnant period. There were still a few fair-sized follicles which were not luteinized, but this was not unexpected in view of the excessive follicle stimulation. Later in metestrus, the ovary was found to be smaller, but the surface was still dotted with corpora (*dog 16*) and many sections revealed only solid masses of corpora lutea (fig. 4). At the end of pseudopregnancy the ovary had regressed to anestrus size, and contained only small regressed corpora.

In view of the marked follicle stimulation, and in an attempt to duplicate the normal estrus more exactly, other tests as to dosage were attempted. Two mature dogs were injected until the first signs of pro-estrus bleeding appeared, and then the hormone was discontinued to see if the cycle would continue. In both cases, pro-estrus of a week's duration and a normal period of estrus followed. This was repeated on an immature dog, with estrus resulting. Therefore, in the second series of dogs this method was employed, and in all but 2 cases estrus was induced, but the total dosage varied considerably (table 1). One experiment was attempted to test the effect of single large doses administered at less frequent intervals. *Dog 24* was given a single injection of 250 R.U., and 4 days later a similar injection. Six days after the first injection, slight vulva swelling and definite bleeding was evident, but after 3 days the bleeding subsided and leucocytes appeared in the smear, although the dog never accepted the male. On the 10th day a second series of injections

was started, using 75 R.U. per day, and continued for 12 days; but estrus was not induced. However, the endometrium exhibited some hypertrophy and the ovary showed follicle stimulation. Possibly longer daily injections would have induced estrus.

The endometrium in estrus was again typically normal, whereas the dogs sacrificed in late pseudopregnancy had a regressed endometrial condition. The ovaries of the mature dogs in estrus exhibited extensive follicle stimulation, although possibly in a smaller number of follicles. The ovaries of the imma-

TABLE 1. EFFECTS OF AN EXTRACT OF MENOPAUSE URINE ('GAMONE'), FOLLUTEIN AND OF PROGYNON-B ON THE ESTRUS REACTIONS OF THE DOG.

Dog	Weight	Period Injected	Total R.U. Injected	Beginning vulva Swelling	Bleeding	Mating	Remarks
			Extract of Menopause Urine 'Gamone'				
	kg.	days		day	days	days	
4	10.3	17	1260	8th	8–18	20–25	Sacrificed 5th day metestrus
7	8.6	20	1260	12th	16–19	21–	Sacrificed 6th day estrus
8	8.2	14	963	12th	—	15–	Sacrificed 3rd day estrus
329	11.0	9	648	9th	10–14	11–19	—
373	12.0	14	990	9th	12–13	14–23	Sacrificed end of metestrus
16	11.6	15	1080	13th	13–16	17–30	Sacrificed 15th day metestrus
21	10.5	6	432	6th	6–14	12–19	Sacrificed 42nd day metestrus
23	7.9	6	432	6th	6–10	13–23	Sacrificed 34th day metestrus
28	7.3	6	572	6th	6– 9	10–15	Sacrificed 5th day estrus
22	11.7	16	1076	14th	15–18	20–25	Sacrificed 2nd day metestrus
24	9.4	2	500	6th	6– 9	—	—
		12	912	—	—	—	Sacrificed 25th day of exp.
30	12.1	21	1550	—	—	—	No effect at 21st day
34	15.2	20	2450	—	—	—	No effect at 22nd day
35	8.3	9	650	14th	little	19–	Sacrificed 1st day estrus
			500 R.U. of Follutein Injected on the 1st Day of Induced Estrus				
36	11.4	17	1450	14th	—	20–27	Sacrificed 31st day metestrus
38	7.6	11	1000	9th	little	15–19	Sacrificed 29th day metestrus
329	11.2	21	2090	24th	24–30	29–38	8 pups 56th day metestrus
			Progynon-B				
5	12.4	15	4500	9th	10–13	19th	Sacrificed 20th day
6	16.6	12	4800	8th	none	13th	Sacrificed 13th day
17	8.7	18	3600	8th	13–16	18–28	Sacrificed 38th day
18	12.9	12	3600	7th	11–13	13th	Sacrificed 14th day

ture dog were not greatly stimulated, and longer injection periods may prove more satisfactory in these cases. *Dog 28,* sacrificed 7 days after the last injection and on a low total dosage, showed luteinization in some follicles, but this would seem to be an effect of the animal's own anterior pituitary rather than one of the extract. In late metestrus the corpora were regressed, and in one case (*21*) were virtually absent on the 42nd day.

Since ovulation points were not found on the ovaries during estrus with either high or low dosage, and the general condition of the ovary indicated that ovulation had not occurred at that time, pregnancy was regarded as the best means of determining whether or not ova were released. Five dogs were tested, and negative results occurred in all cases. Ovulation may have been

prevented, since overstimulation of the follicles during follicular development has been suggested to incapacitate the follicle for ovulation (24).

Three experiments have been performed in an effort to produce ovulation in an induced cycle by injecting 500 R.U. of follutein intraperitoneally on the first day of estrus. These dogs were bred, and in one case the dog delivered 8 pups on the 56th day, all of which were weaned and were normal in every respect. Thus the combination of 'gamone' and follutein induced estrus and ovulation in the dog comparable in every respect to the spontaneous cycle. Further experiments will be necessary to determine definitely whether ovulation always occurs following administration of follutein. Neither of the other 2 dogs was sacrificed in estrus to observe possible ovulation points, but was carried to mid-metestrus, at which time laparotomies were performed and, since they were not pregnant, the animals were sacrificed. The ovaries of these dogs were large, consisting almost entirely of corpora lutea, which appeared active. The corpora were much more extensive than with 'gamone' alone, and seemed more capable of maintaining a complete pseudopregnant period.

Dog 329 presented an interesting study. In February, 1936, estrus was induced in this animal with menopause urine extract. The following December a spontaneous estrus occurred, and 2 months later, at the end of pseudopregnancy, estrus was induced for a second time with this extract and the dog tested for ovulation by pregnancy. This being a negative case, the dog was subjected to a third series of injections 3 months after the previously induced cycle. Estrus resulted for the third time, and on the first day of estrus follutein was injected. The dog became pregnant and delivered. The follicle-stimulating action of the extract, therefore, does not exert a deleterious effect on the ovary.

Experimental induction of estrus with progynon-B. The induction of estrus in the dog solely by means of injections of estrone preparations has been claimed and denied by various investigators. Some of these failed to observe any external indications of estrus in the normal dog following administration of this hormone (25, 26, 27, *dog D3*). On the other hand, vulva swelling and pro-estrus bleeding have been observed, although mating was not tested (27, *dog D6,* 28). Complete induction of estrus in the normal dog has been reported (5, 6, 7). In the castrate, pro-estrus bleeding has resulted from injections of estrone (28, 29), and also true estrus as determined by mating (6, 27). In most of these studies, the extract used was not nearly as well purified and potent as the estrone preparations available at the present time, and therefore it was deemed of value to repeat these experiments and compare this induced estrus with the normal cycle.

Three normal dogs and one castrate were injected with progynon-B, in oil. Vulva swelling and mating were induced in every case. Swelling of the vulva was first, evident between the seventh and ninth days and, at the time of estrus, was comparable in size to that of normal estrus. One dog maintained maximal swelling for 20 days after the last injection, at which time the animal was sacrificed. Definite macroscopic hemorrhagic discharge resulted in the 3 normal dogs for 2 to 3 days, which is a somewhat shorter period than the normal. Since most of the uterus had been removed, no bleeding occurred in

the castrate. Mating with normal males was tested, and copulation resulted in all cases. Injections were discontinued on the first day of estrus, but one dog continued to mate for 10 days, showing a normal estrus period. The other dogs were sacrificed following the first successful mating.

Vaginal smears were taken daily and compared to those of the normal cycle. The smear of *dog 17* remained cornified during the period the animal accepted the male. This was followed by a great increase in leucocytes comparable to the beginning of metestrus in the control. However, 8 days after the first appearance of leucocytes, the smear had reverted to an anestrus condition.

The uterine endometrium in estrus was hypertrophied with an increase in the uterine glands. The dogs receiving the largest total dosage exhibited a slightly more advanced development than the other two animals, but all were of an early estrus type. At the end of estrus, further endometrial changes in the glandular complexity take place in control and 'gamone' dogs but, in spite of carrying one progynon-B dog until 10 days after estrus, the endometrium had progressed no further than the early estrus condition (fig. 5).

Estrone injected into immature dogs has been found to cause regression of the ovary to half size (30), but the effect on the ovary of the mature dog has not been considered. The ovaries of the 3 normal dogs injected with progynon-B were of anestrus size, averaging 0.4 gm. in weight, which compares with the normal anestrus ovary. However, many more experiments would be necessary to consider a possible regression resulting from injections of this hormone. The general condition of the ovary exhibited only primary and growing follicles in all cases (fig. 6), and therefore the ovary was seemingly not effected. It is obvious that a metestrus period could not follow this induced estrus.

DISCUSSION

The results of the experiments with an extract of menopause urine 'gamone' indicated that its action on the normal dog was comparable to the F.S.H. of the anterior pituitary. The ovaries exhibited excessive follicle stimulation in the absence of luteinization, regardless of dosage, although in a few follicles there was a slight thickening of the granulosa. The ovaries were found to be as much as 10 times normal anestrus size, but the follicles contained normal ova. In comparison to as many as 39 in a single ovary of the mature dog, only a few follicles were greatly stimulated in the immature dog.

Mature dogs, in pseudopregnancy and anestrum, as well as immature dogs respond to the extract, and in these experiments estrus was successfully induced in 14 of the 16 cases tested. The dogs were injected with 75 R.U. daily until the first day of estrus, but later it was found that a cycle accentuated to the time of pro-estrus would continue on into estrus. In these cases the pro-estrus period was of more normal duration. That smaller daily dosages prove more effective than a comparable total dosage administered in 2 injections was tested in one case.

Not only are all the external manifestations of true estrus induced, namely,

vulva swelling, pro-estrus bleeding and mating, but also endometrial hypertrophy and the typical vaginal smear changes. In addition, this estrus is followed by a normal pseudopregnant period as determined by the glandular complexity of the endometrium and the corpora lutea of the ovary.

Whereas various estrone preparations have been used to induce estrus in the dog, progynon-B had previously not been tested. This hormone induced vulva swelling, hemorrhagic discharge, endometrial hypertrophy and mating, as well as changes in the vaginal smears indicative of estrus. Too short an injection period, or the lack of sufficient potency in the various follicular ovarian hormones used, or both, may possibly account for the failure to obtain all of these results in other instances.

The cycle induced by progynon-B fails to duplicate the normal in all respects because it is not followed by a metestrus phase. This was evident by the premature return of the smear to an anestrus type, the failure of the endometrium to progress beyond the early estrus type and the fact that the ovary is not stimulated but remains in an anestrus condition. This likewise explains why Grollman (7) could not demonstrate a prolonged life span with dogs not receiving cortical hormone, and which had been adrenalectomized during estrus induced with theelin, and it also lends support to his assumption that this cycle fails in some respect to duplicate the normal. However, the fact that a pseudopregnant period follows estrus induced with menopause urine extract in the normal dog is supported by the work on the adrenalectomized animal (1, 2).

Using pregnancy as the criterion to determine ovulation in the cycles induced with menopause urine extract, 5 dogs failed, thus suggesting that ovulation did not result. In an effort to duplicate the spontaneous cycle, 3 dogs were injected with follutein on the first day of estrus, and one of these delivered 8 pups.

SUMMARY

All external manifestations of estrus, namely, vulva swelling, bleeding and mating, as well as endometrial hypertrophy may be induced in the normal dog with either an extract of menopause urine or progynon-B. Induction of estrus with menopause urine extract was successful with mature dogs in anestrus or metestrus, as well as with immature dogs in 14 of the 16 cases tested. This extract produced pure follicle stimulation comparable to the F.S.H. of the anterior pituitary, whereas progynon-B exerted no obvious effect on the ovary. A cycle accelerated to pro-estrus with menopause urine extract will continue through estrus quite normally, and is followed by a pseudopregnant period, thus duplicating the spontaneous cycle. A pseudopregnant period does not follow estrus induced with progynon-B. Ovulation was not observed with menopause urine extract alone, but injection of follutein on the first day of estrus resulted in pregnancy in 1 of 3 cases.

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REFERENCES

1. Swingle, W. W., W. M. Parkins, A. R. Taylor and J. A. Morrell: Proc. Soc. Exper. Biol. & Med. **41**:94. 1936.
2. Swingle, W. W., W. M. Parkins, A. R. Taylor, H. W. Hays and J. A. Morrell: Am. J. Physiol. **119**:675. 1937.
3. Rogoff, J. M., and G. N. Stewart: Am. J. Physiol. **86**:20. 1928.
4. Pfiffner, J. J., W. W. Swingle and H. M. Vars: J. Biol. Chem. **104**:701. 1934.
5. Papanicolaou, G. N., and N. F. Blau: Anat. Rec. **35**:47 (abst.). 1927.
6. Kunde, M. M., F. E. D'Amour, A. J. Carlson and R. G. Gustavson: Am. J. Physiol. **95**:630. 1930.
7. Grollman, A.: The Adrenals. Williams & Wilkins, Baltimore, 1936.
8. Evans, H. M., and H. H. Cole: Mem. Univ. California **9**:65. 1931.
9. Zondeck, B.: Klin. Wchnschr. **9**:393. 1930.
10. Hamburger, C.: Acta. path. et microbiol. Scandinav. **17**:1(Suppl.). 1933.
11. Leonard, S. L., and P. E. Smith: Am. J. Physiol. **108**:22. 1934.
12. Smith, P. E., E. T. Engle and H. H. Tyndale: Proc. Soc. Exper. Biol. & Med. **31**:745. 1934.
13. Smith, P. E., and E. T. Engle: J. Pediat. **5**:163. 1934.
14. Selye, H., J. P. Collip and D. L. Thomson: Proc. Soc. Exper. Biol. & Med. **32**:800. 1935.
15. Engle, E. T. (unpublished; cited by P. E. Smith): J.A.M.A. **104**:553. 1935.
16. Osterreicher, W.: Klin. Wchnschr. **11**:813. 1932, *ibid.*, **14**:1570. 1935.
17. Lassen, H. C. A., and E. Bandstrup: Acta. obst. et gynec. Scandinav. **14**:89. 1934.
18. Evans, H. M., and M. E. Simpson: Proc. Soc. Exper. Biol. & Med. **32**:1046. 1935.
19. Lipschutz, A.: Endocrinology **19**:42. 1935.
20. Frank, R. T., U. J. Salmon and R. Freidman: Proc. Soc. Exper. Biol. & Med. **32**:1666. 1935.
21. Salmon, U. J., and R. T. Frank: Proc. Soc. Exper. Biol. & Med. **34**:463. 1936.
22. Frank, R. T., and U. J. Salmon: Proc. Soc. Exper. Biol. & Med. **35**:493. 1936.
23. Tyndale, H. H., and L. Levin: Am. J. Physiol. **120**:486. 1937.
24. Hisaw, F. L., R. O. Greep and H. L. Fevold: Anat. Rec. **61**:24 (Suppl.). 1935.
25. Asdell, S. A., and F. A. H. Marshall: Proc. Roy. Soc. Med. **101B**:185. 1927.
26. Koch, W.: München. Tier. Wchnschr. **84**:551. 1933.
27. Robson, J. M., and W. R. Henderson: Proc. Roy. Soc. Med. **120B**:1. 1936.
28. Mizuno, T.: Jap. J. Med. Sciences, Pharm. **2**:1. 1927.
29. Meyer, R. K., and S. Saiki: Proc. Soc. Exper. Biol. & Med. **29**:301. 1931.
30. Kunde, M. M., F. E. D'Amour, R. G. Gustavson and A. J. Carlson: Proc. Soc. Exper. Biol. & Med. **28**:122. 1930, *ibid.*, Am. J. Physiol. **96**:677. 1931.

INHIBITION OF GONADOTROPIC ACTIVITY BY SEX HORMONES IN PARABIOTIC RATS

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There seems to be no doubt that castration in the rat results in an increase in the gonad-stimulating capacity of the hypophysis, and it is also generally accepted that estrogenic substances depress the gonadotropic power of the pituitary (1). Moore and Price have shown that bull testis extracts (2) and androsterone (3) injected into young male rats cause testicular damage. They attributed this injurious effect to a lowering of pituitary gonad-stimulating hormone, an idea supported by the fact that implantation studies showed the pituitaries of androsterone-injected rats to be reduced in gonadotropic potency. Of special interest are recent reports (4, 5) that ovarian weights in immature parabiotic rats could be used as a quantitative measure of hypophyseal gonadotropic secretion of castrated parabionts which were untreated or treated with estrone or androgenic substances. Our own observation (6, 7) on paraboitic rats suggested the possibility that the testicular response of hypophysectomized rats might serve as a qualitative indicator of the hypophyseal gonadokinetic activity of a castrated parabiont.

Since these suggestive observations have been given in detailed form elsewhere, it will be necessary here only to review them briefly. *A.* Provided that sufficient time has been allowed for reactions to manifest themselves, the testes of hypophysectomized rats parabiotically joined with gonadectomized parabionts (irrespective of sex) have been invariably maintained in a completely normal condition, or, if atrophic, restored to one. *B.* The gonadotropic hormonal complex of the pituitary of a castrated rat causes parallel stimulation of both the interstitial and reproductive cells of the testes of a hypophysectomized twin. A selective gametokinetic response has never been observed in such cases. *C.* Flushing and turgescence of the scrotum may be regarded as very sensitive qualitative indicators of interstitial cell activity. Such scrotal reactions may also serve as rough quantitative indicators of male hormone. *D.* Within certain limits, grossly perceptible increase in the size of the testis may be attributed chiefly to gametogenic activity. This is especially true in instances when the testes, on palpation, are nearly normal in size. The correlation between testis size and tubular growth is readily understandable, when it is realized that the interstitial tissue at best constitutes but a very small fraction of the total volume of the testis.

The appearance of the scrotum and the size of the testis in the hypophysectomized parabiotic rat may therefore be considered, respectively, as differential manifestations of interstitial cell activity and germ cell activity. If it be true that each of these types of activity is controlled by a separate pituitary

hormone (8, 9), then the condition of the scrotum or the size of the testis in a hypophysectomized twin would reflect the nature of the pituitary gonadokinetic hormone emanating from the pituitary of a partner. On this basis, growth of the scrotum, being controlled by hormone from the interstitial cells, would indicate release of so-called luteinizing hormone (8, 9) or of interstitial cell-stimulating hormone (10). Similarly, the size of the testis, being a sign of gametogenic activity, would indicate the release of a gametokinetic principle or follicle-stimulating hormone (8, 9).

Testicular stimulation in hypophysectomized rats has to some extent proved to be reliable as a quantitative indicator of gonadokinetic hormone received from the pituitary of a partner, as well as a qualitative indicator. By this means it has been possible to determine the effects of androgenic and estrogenic substances on the gonadotropic activity of the pituitaries of castrated and normal rats. The effects observed under these conditions form the basis of this report. It must be remembered that the results to be described are preliminary in the sense that they are based on reconnaissance experiments. All of the pairs upon which these data are based are still alive and in good health, and will be used repeatedly in an attempt to discover the smallest dose of substance effective in checking gonadotropic activity of the pituitary.

MATERIALS AND METHODS

Two kinds of rats were used; albino, and black-eyed tan-hooded. The latter strain was reared in our own laboratory and has been very closely inbred. No anatomical or physiological characteristics peculiar to either strain have ever been observed, and both strains have always responded quite similarly under corresponding experimental conditions.

The animals joined in parabiosis were in most instances litter mates whose ages ranged from 50 to 120 days at the time of operation. Numerous injected and uninjected non-parabiotic control rats were under observation during the course of this investigation. Data obtained on these unjoined controls in no way conflicted with the findings on parabiotic rats, and hence will be omitted from this report.

As we have introduced some helpful modifications into the technic of parabiosis which we formerly used (6), it seems advisable to describe briefly this improved procedure. The rats to be joined parabiotically were anesthetized with ether and laid on a table facing each other, one with its right side up and the other with its left side up. Clippers were used to remove the hair from each of these sides, which were then washed with 70 per cent alcohol. The clipped skin was grasped with forceps so that it formed a single fold extending from the region of the thigh to about 1/2 to 3/4 of an inch caudal to the ear. The crest of this skin-fold was cut off with scissors, resulting in the excision of a piece of skin whose length varied with the length of the animal, and whose width was about 1/4 inch. A short incision (1/2 to 3/4 inch) was then made through the abdominal musculature and peritoneum in each animal. The edges of these latter wounds were approximated like 2 pairs of closed lips and fixed thus with interrupted sutures of silk, resulting in an imperforate union of the lips of the wound. While the peritoneal cavities of the 2 animals

thus remained separate, an area was provided in which adequate anastomosis of the capillaries of the parabionts could occur. A single suture was taken through the subcutaneous connective tissues at the cranial and caudal ends of the incisions. These points were subjected to great strain, and sutures through the subcutaneous tissues were found greatly to relieve strain on the skin. Metal wound clips (11-16 mm.) were used to bring together the edges of the skin. Starting about 3 or 4 days after the operation, one or two clips were removed every day or so, until none remained. Care had to be exercised in this respect, as it was found that too early removal of a clip from a point subjected to much strain would lead to tearing apart of the animals. On the other hand, clips left in the skin too long sometimes caused infection or tears in the skin which resulted in imperfect healing.

At some time during the process of healing (1 to 2 weeks) or somewhat later, a parabiotic animal may go through a critical period. In this stage the animal may present one or all of the symptoms described by Selye (11) under the term 'alarm reaction.' Many fatalities occur in this critical period. For this reason we usually join 2 normal rats in parabiosis, and wait 2 or 3 weeks before performing any other operation on them.

After a pair of normal animals has successfully passed through the critical period described, one of the members of the pair is hypophysectomized. Atrophy of the gonads may be retarded, but in our experience it has invariably occurred eventually. This is an important stage, as it affords evidence of the completeness of hypophysectomy. In view of the fact that it has seemed desirable not to sacrifice the pairs on which we are reporting, the critical proof of complete hypophysectomy afforded only by histological examination of the sellar contents is necessarily lacking. However, cessation of growth and loss in body weight in a hypophysectomized parabiont, together with obviously retarded anteroposterior development of the skull (12) and gonadal atrophy, may be regarded as fairly reasonable assurance of complete pituitary ablation. Additional evidence of the completeness of hypophysectomy is furnished at the time of operation. The technic of hypophysectomy is essentially that described by Thompson (13), involving aspiration of the gland into a glass tube. The pieces of pituitary drawn into the suction tube may be recovered and pieced together to form the complete hypophysis, if the operation has been successful.

If the normal partner of a hypophysectomized twin showing testicular atrophy is castrated, the scrotum of the hypophysectomized animal becomes flushed and edematous within 2 or 3 days. This scrotal flushing and turgescence may be accompanied, or shortly followed, by descent of the atrophic testes into the scrotum. Under the influence of a gonadotropic secretion from the pituitary of the castrate, development of the testes and scrotum in the hypophysectomized twin will continue until the entire genital tract is restored to a normal condition (6, 7).

The substances to be injected were dissolved in sesame oil and administered subcutaneously[1]. All other pertinent data will be found in the tables.

[1] For the crystalline androgenic and estrogenic substances employed in this work we are indebted to Dr. Erwin Schwenk of the Schering Corporation.

TABLE 1. CASTRATED MALE RAT (85c)+HYPOPHYSECTOMIZED MALE RAT (85h)

April 28, 1937. 85h was hypophysectomized, 41 days before parabiosis.
April 15, 1937. 85c was castrated, 54 days before parabiosis.
June 8, 1937. Parabiosis when animals were about 3 months old.
June 16, 1937. Definite stimulation of scrotum of 85h was seen.
June 22, 1937. Testes and scrotum of 85h were well stimulated.

Time	Treatment in Castrated Twin				Reaction of Testes and Scrotum in Hypophysectomized Twin
	Substance	Daily Dose: Amount (gamma)	Daily Dose: Oil (cc.)	Number of Injections	
June 22 to July 2[1]	Testosterone propionate = TP	3000	1/4	7	Progressive atrophy.
July 3 to Aug. 2	No treatment				Gradual return toward normal condition. See fig. 1 (July 20).
Aug. 3–12	TP	3000	1/4	10	Gradual degeneration which was quite definite by Aug. 18. See fig. 2 (Aug. 18).
Aug. 13–17[2]	TP	1500	1/8	2	
Aug. 18–23	TP	250	1/4	6	Atrophy pronounced.
Aug. 24–31	TP	125	1/4	8	Marked atrophy.
Sept. 1–3	TP	63	1/8	3	Continued atrophy. See fig. 3 (Sept. 8).
Sept. 4–8	TP	94	3/16	5	
Sept. 9–12	TP	50	1/10	4	
Sept. 13–19	No treatment				Atrophy persisted.
Sept. 20–28	No treatment				Gradual growth. See fig. 4 and 5 (Sept. 20 and 28)

[1] No injections given June 27, 29, and July 1, 2.
[2] No injections given Aug. 13, 15, 17.

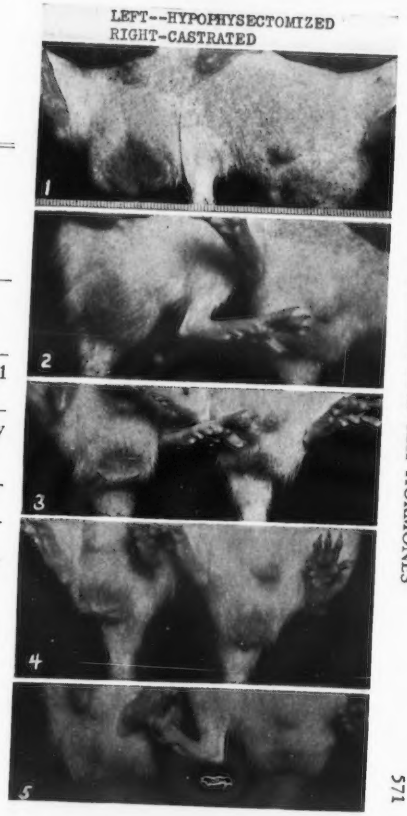


TABLE 2. CASTRATED MALE RAT (120c)+HYPOPHYSECTOMIZED MALE RAT (120h)[1]

May 25, 1937. Parabiosis of litter mates aged 54 days.
June 3, 1937. 120h was hypophysectomized, 9 days after parabiosis. Regression of scrotum and testes was not complete until about 7 weeks after hypophysectomy.
July 22, 1937. 120c was castrated, 58 days after parabiosis.

Time	Treatment in castrated male				Reaction of Scrotum and Testes in Hypophysectomized Male
	Substance	Daily Dose		Number of Injections	
		Amount (gamma)	Oil (cc.)		
July 22 to Aug. 17	No treatment				Gradual return toward normal condition. See fig. 6 (July 30).
Aug. 18–23	Estradiol	5	1/20	6	No apparent change.
Aug. 24–25	Estradiol	25	1/4	2	
Aug. 26–28	Estradiol	25	1/4	3	A slight regression of scrotum seemed to occur Aug. 26–28, but this was not permanent. Scrotum and testes appeared normal Sept. 3.
Aug. 29–30	Estradiol	50	1/2	2	
Aug. 31 to Sept. 3	Estradiol	100	1/4	4	
Sept. 4–6	No treatment				No change.
Sept. 7 to Sept. 19	Estrone	100	1/4	13	No marked change at first. See fig. 7 (Sept. 8). After 10 injections atrophy occurred quickly and definitely. See fig. 8 (Sept. 20).
Sept. 20 to Sept. 28	No treatment				Regression persisted for about 3 days and then there was a gradual return toward normal condition. See fig. 9 (Sept. 28).

[1] 120h is suspected of being incompletely hypophysectomized.

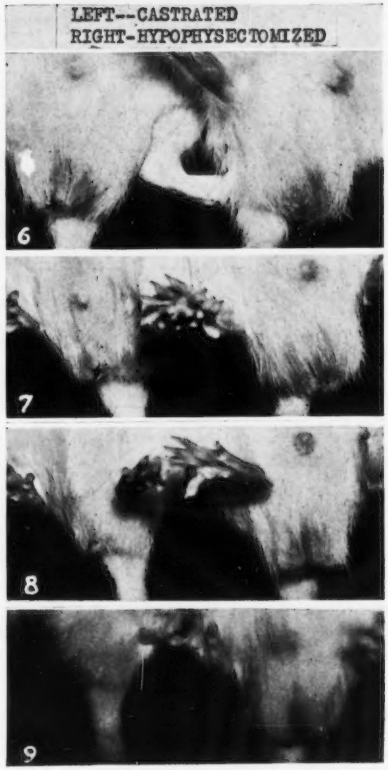
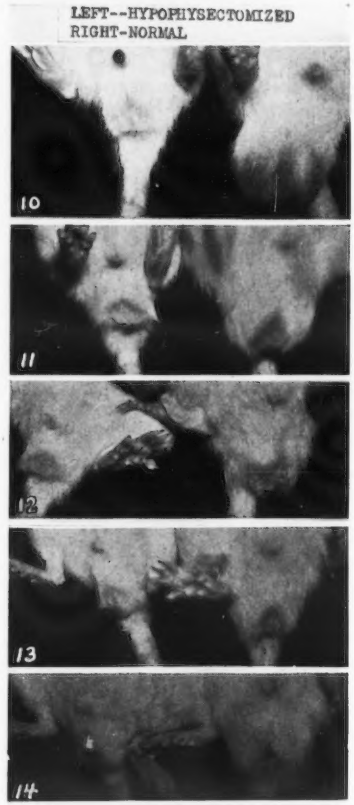


TABLE 3. NORMAL MALE RAT (123n)+HYPOPHYSECTOMIZED MALE RAT (123h).

May 28, 1937. Parabiosis of litter mates aged 54 days.
July 19, 1937. 123h was hypophysectomized, 52 days after parabiosis. Regression of testes and scrotum was not at all retarded; it was thoroughly complete by Aug. 18 (fig. 10).

Time	Treatment in normal male				Reaction of Testes and Scrotum in Normal and Hypophysectomized Twins
	Substance	Daily Dose: Amount (gamma)	Daily Dose: Oil (cc.)	Number of Injections	
Aug. 18–23	Testosterone propionate	250	1/8	6	Normal twin. Except for some thickening of scrotal wall, there was no apparent change.
Aug. 24–25	Testosterone propionate	500	1/4	2	
Aug. 26–28	Testosterone propionate	750	1/2	3	Hypophysectomized twin. No effect.
Aug. 29–31	Testosterone propionate	1000	1/2	3	
Sept. 1–6	No treatment				No change in either animal.
Sept. 7–15	Estrone	100	1/4	9	Normal twin. Regression began after 4 injections and was very obvious after 8. Compare fig. 11 (Sept. 8) with fig. 12 (Sept. 15). Hypophysectomized twin. No effect.
Sept. 16–28	No treatment				Normal twin. Atrophy persisted for about 6 days: see fig. 13 (Sept. 20). Stimulation was not definite until Sept. 28 (fig. 14). Hypophysectomized twin. No effect.



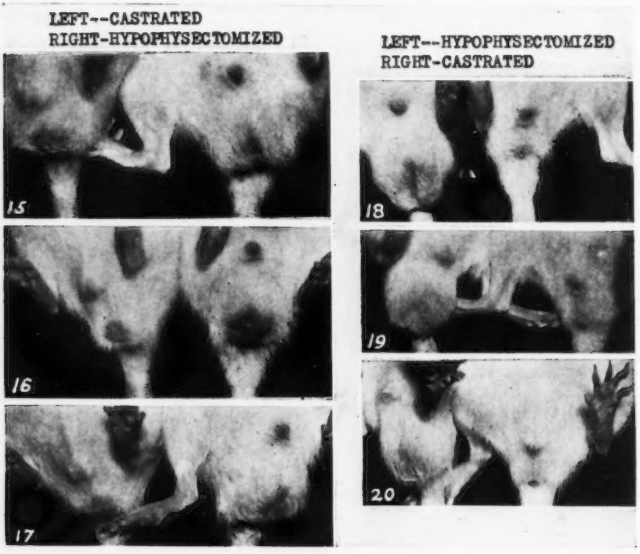


TABLE 4. CASTRATED MALE RAT (134c)+HYPOPHYSECTOMIZED MALE RAT (134h).

June 8, 1937. Parabiosis at age 60 days.
July 22, 1937. 134h was hypophysectomized, 44 days after parabiosis. Regression of testes and scrotum was not complete until 4 weeks after pituitary ablation.
Aug. 18, 1937. 134c was castrated, 71 days after parabiosis.

Time	Treatment in castrated male				Reaction of Testes and Scrotum in Hypophysectomized Male
	Substance	Daily Dose		Number of Injections	
		Amount (gamma)	Oil (cc.)		
Aug. 18–20	No treatment				Scrotum became flushed and turgescent.
Aug. 21–22	Estrone	5	1/20	2	Although possibly somewhat retarded, growth continued.
Aug. 23	Estrone	10	1/10	1	
Aug. 24–27	Estrone	25	1/4	4	
Aug. 28–30	Estrone	50	1/2	3	
Aug. 31 to Sept. 3	Estrone	100	1/4	4	Very suddenly scrotum lost color. Atrophy progressed rapidly. See fig. 15 (Sept. 8).
Sept. 4–6	Estrone	75	3/16	3	
Sept. 7–11	Estrone	50	1/8	5	
Sept. 12	Estrone	25	1/16	1	
Sept. 13–18	No treatment				No apparent signs of stimulation.
Sept. 19–28	No treatment				Stimulation began: see fig. 16 (Sept. 20). Development was rapid: see fig. 17 (Sept. 28).

TABLE 5. SPAYED FEMALE RAT (148s)+HYPOPHYSECTOMIZED MALE RAT (148h).

Aug. 6, 1937. Parabiosis at age 53 days.
Aug. 26, 1937. Female was spayed and male was hypophysectomized, 20 days after parabiosis.

Time	Treatment in Spayed Female				Vaginal Smear in Spayed Female	Reaction of Testes and Scrotum in Hypophysectomized Male
	Substance	Daily Dose		Number of Injections		
		Amount (gamma)	Oil (cc.)			
Aug. 27 to Sept. 5	No treatment				Di-estrous, occasionally mixed	Genitalia perfectly maintained. See fig. 18 (Sept. 8).
Sept. 6–20	Estrone	5	1/20	15	Cornified[1]	No definite change.
Sept. 21–25	Estrone	25	1/16	5	Cornified[2]	Signs of regression. See fig. 19, 20 (Sept. 20 and 28).
Sept. 26	Estrone	15	3/20	1	Cornified	Atrophy was progressing.

[1] Some leucocytes in smear Sept. 14 and 20.
[2] Some leucocytes in smear Sept. 21 and 22.

RESULTS

Little explanation need be added to that furnished by the tables. Undue emphasis should not be placed upon the dosages used, as they were empirically selected. Analysis of the data in tables 1, 2, 4, and 5 yields at least the following information.

An increase in gonadokinetic activity occurred in the pituitaries of adult male or female rats following castration, and caused testicular stimulation in hypophysectomized male twins. Daily injections of testosterone propionate (3000 to 50 γ) into the castrated male twin prevented hypophyseal gonadotropic activity (table 1). It might be mentioned, too, that the scrotum of the castrated partner showed varying degrees of flushing and turgescence in response to the injected hormone. Daily injections of α-estradiol (5 to 100 γ) into a castrated male twin failed to check hypophyseal gonadotropic hyperactivity, but estrone at a level of 100 γ did inhibit this hyperactivity (table 2). It should be noted that the test twin in this instance was believed to be incompletely hypophysectomized. Increased gonadotropic activity of the pituitary in another castrated male parabiont was prevented by daily injections of estrone ranging from 100 to 25 γ (table 4). Daily doses of estrone (25 to 5 γ) inhibited gonadotropic hyperactivity in a young adult spayed female parabiont joined with a hypophysectomized male (table 5).

Daily injections of testosterone propionate (250 to 1000 γ) into a normal male parabiont joined with a hypophysectomized male twin seemed to induce no significant change in the external genitalia of either partner. However, 9 daily injections of estrone at a level of 100 γ injected into the normal twin caused marked atrophy of its scrotum and testes. This effect was not accompanied by any gross change in the genitalia of the hypophysectomized male twin (table 3).

Figure 21 presents graphically some data on the vaginal changes in a

hypophysectomized female rat joined with a castrated male twin. Under these conditions the hypophysectomized female fell into a state of continuous vaginal estrus, as described by Witschi and Levine (14). It will be seen in the figure that injections into the castrated male of either estrone (5 to 15 γ) or testosterone propionate (3000 γ once a week) more or less interrupted the continuous estrus in the hypophysectomized female. The interruptions following the single large doses of testosterone propionate occurred with striking regularity. Such rhythmic changes in the vagina were apparently modified estrous cycles. As the condition of the ovaries in the hypophysectomized female was not known at any time, it will be difficult fully to interpret these findings.

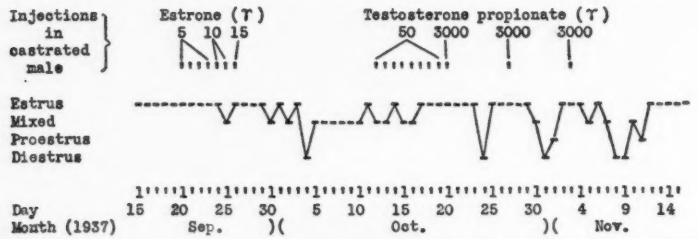


Fig. 21. Daily vaginal smear record of hypophysectomized female rat joined with castrated male.

DISCUSSION

It seems justifiable to conclude that testosterone propionate injected into castrated males, or that estrone injected into castrated males or females, largely or completely suppressed the gonadokinetic activity of their pituitary glands. In all probability the suppression was complete in those instances where suspension of treatment was not followed in 2 or 3 days by renewed testicular stimulation in the hypophysectomized twin. This is all the time ordinarily required for the scrotal reaction, which has been described, to manifest itself. It will be interesting in future studies to correlate physiological effects with histological and quantitative findings on the pituitary of the castrated parabiont, and on the sexual organs of the castrated and hypophysectomized twins.

As far as it was possible to determine by this method, pituitary gonadotropic inhibition in castrated partners was invariably followed by parallel degeneration of both the interstitial and spermatogenic elements of the testes of the hypophysectomized test object. If both these elements are, as previously suggested (6, 15), dependent upon only one pituitary gonadokinetic hormone, then complete gonadal atrophy can be accounted for on the basis of suppression of this one factor. Assuming that the gametogenic and endocrine functions of the testes are controlled by separate gonad-stimulating hormones, it follows that both these principles are elaborated and released by the pituitary of castrated male or female rats, and that both are simultaneously inhibited by male or female hormone. In such cases, as well as in the case where a normal twin was injected with androgenic or estrogenic substances, no evidence of interstitial cell activity (scrotal reaction) which might be attributed to release of a pituitary principle was ever detected in the hypophysectomized test

animal. The scrota of the test rats were examined carefully several times daily with this point in mind, as the theory has been advanced that estrone in some way brings about a release of a luteinizing hormone from the pituitaries of female rats (16, 17, 18).

It is interesting to note that injections of testosterone propionate into a castrated male joined with a hypophysectomized female exhibiting continuous cornification of the vagina caused the vaginal epithelium of this female to undergo rhythmic changes resembling those of the estrous cycle of normal female rats. In view of the fact that sex hormones have been found to effect a suppression of gonadokinetic hormone in male and female castrates, it seems quite possible that the cyclic vaginal changes in the hypophysectomized female were caused by different levels of follicular hormone from her ovaries under the influence of different quantities of a single hypophyseal gonadotropic hormone from the castrated partner. This conclusion, while likely, is not warranted at present, as we have no data on the condition of the ovaries in the hypophysectomized test female.

SUMMARY

The testes of hypophysectomized male rats parabiotically joined with castrated male or female rats invariably show parallel stimulation of the spermatogenic and interstitial elements. Under such conditions it has been found that *a,* within certain limits, grossly discernible increase in the size of the testes reflects chiefly tubular growth and development, whereas marked decrease in testicular size indicates predominantly degeneration of gametogenic elements, and *b,* the physiological state of the interstitial cells to some extent is mirrored by the condition of the scrotum.

Injection of varying doses of testosterone propionate or of estrone into castrated male or female parabiotic rats depressed the gonadotropic hormone of their pituitary glands to such an extent that stimulation of the testes of their hypophysectomized twins was prevented or stopped. When testicular stimulation was stopped under these conditions, parallel degeneration of the tubular and interstitial cells occurred. This may be interpreted in terms of suppression of one or two hypophyseal gonad-stimulating hormones.

In a pair consisting of a castrated male and a hypophysectomized female, the female fell into a state of continuous estrus, as revealed by daily vaginal smears showing only cornified cells. Injections of testosterone propionate, and to a lesser extent estrone, into a castrated partner led to the appearance of cyclic variations in the vaginal smear of the hypophysectomized twin which were similar to those characteristic of normal estrous cycles. In the absence of knowledge of accompanying ovarian changes in this case, only a tentative interpretation of the data was ventured. It is suggested that the cyclic vaginal changes were caused by fluctuations in the output of follicular hormone from the ovaries of the hypophysectomized female, depending upon the quantity of a single gonadotropic hormone released by the pituitary of the castrated male.

The authors wish to express their gratitude to Dr. William E. Lower for the continued interest he has taken in this work.

REFERENCES

1. Allen, E.: Sex and Internal Secretions, Chap. XVII, Williams & Wilkins, Baltimore, 1932.
2. Moore, C. R., and D. Price: Am. J. Anat. **50**:1. 1932.
3. Moore, C. R., and D. Price: Endocrinology **21**:313. 1937.
4. Meyer, R. K., and R. Hertz: Am. J. Physiol. **120**:232. 1937.
5. Hertz, R., and R. K. Meyer: Endocrinology **21**:756. 1937.
6. Cutuly, E., D. R. McCullagh and E. Cutuly: Endocrinology **21**:241. 1937.
7. Cutuly, E., and E. C. Cutuly: **37**:477. 1937. Proc. Soc. Exper. Biol. & Med.
8. Greep, R. O., H. L. Fevold and F. L. Hisaw: Anat. Rec. **65**:261. 1936.
9. Greep, R. O., and H. L. Fevold: Endocrinology **21**:611. 1937.
10. Evans, H. M., K. Kopi, M. E. Simpson, R. I. Pencharz and D. H. Wonder: Univ. California Publ. Anat. **1**:225. 1936.
11. Selye, H.: Endocrinology **21**:169. 1937.
12. Mortimer, H.: Radiology **28**:5. 1937.
13. Witschi, E., and W. T. Levine: Proc. Soc. Exper. Biol. & Med. **32**:101. 1934.
14. Thompson, W. K.: Endocrinology **16**:357. 1932.
15. Cutuly, E., D. R. McCullagh and E. C. Cutuly: Am. J. Physiol. **119**:121. 1937.
16. Fevold, H. L., F. L. Hisaw and R. Greep: Am. J. Physiol. **114**:508. 1935.
17. Witschi, E., and C. A. Pfeiffer: Anat. Rec. **64**:85. 1935.
18. Pfeiffer, C. A.: Anat. Rec. **67**:159. 1937.

ELECTRICAL EXCITABILITY (CHRONAXIE) OF THE RAT'S UTERUS, THROUGHOUT THE ESTRUS CYCLE[1]

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Although the motility of the uterus has been the object of considerable investigation (1), the literature contains little reference to any quantitative observations on the excitability of this organ (2, 3). In view of the important influence of the endocrine balance on uterine motility, during the estrus cycle in animals and the menstrual cycle in humans, it seemed likely that the excitability of the uterus would be subject to similar influence. Moreover, the measurement of excitability as an index of hormonal status would be preferable to the recording of motility, since the former property is amenable to more exact determination in terms of electrical excitability or chronaxie (4, 5, 6, 7). It therefore seemed worth while to study the chronaxie of the rat's uterus throughout the normal estrus cycle and during various experimentally induced hormonal imbalances.

The present report is confined to the results obtained during the normal estrus cycle, and demonstrates a cyclic variation in the electrical excitability of the uterus co-incident with the normal estrus cycle.

METHODS

The measurement of the chronaxie of a tissue involves the observation of its response to an electrical stimulus of known strength and duration. The smallest voltage capable of evoking a perceptible response from the tissue is known as the rheobase. To measure the chronaxie, a current of twice the rheobase is used, and the shortest time of action of this current necessary to produce just the minimum response is determined. The unit of chronaxie is therefore a time unit, the σ (sigma $= 1/1000$ second.) The shorter the chronaxie, the greater the excitability of the tissue.

The apparatus was built by the author and consisted of a system of resistors (5) with an average value of about 11,000 ohms, through which a galvanic current (0-250 volts) was conducted to the tissue being tested. Up to 100 σ the duration of the electrical stimulus was determined by the discharge of plate condensers (in this arrangement 1 MF $= 4\,\sigma$). Between 100 and 5000 σ a mechanical rheotome was used.

The rats used in these experiments were carefully followed by the vaginal smear method for an adequate period before they were sacrificed to determine

[1] This work was begun at the Maternity Hospital of the University of Berne, Switzerland (Professor Hans Guggisberg). At that time, the author did not realize the existence of two types of uterine contraction. For this reason, the earlier results had to be largely discarded and the work started afresh under the direction of Dr. Samuel Soskin, Michael Reese Hospital.

[2] Aided by the A. B. Kuppenheimer Fund.

the chronaxie of the uterus at any particular point in the estrus cycle. The status as indicated by the vaginal smears was checked after each experiment by postmortem examination of other organs. On the day of the experiment, the rat was killed by a blow on the head and the uterus quickly but gently removed. Each uterine horn was suspended from silver chloride electrodes in a separate glass container of freshly prepared Tyrode's solution, maintained at a constant temperature of 36°C. (normal body temperature of the rat). The ovarian end of the uterine horn to be tested was connected to the anode wire, and this same wire attached to a counter-balanced lever, which recorded

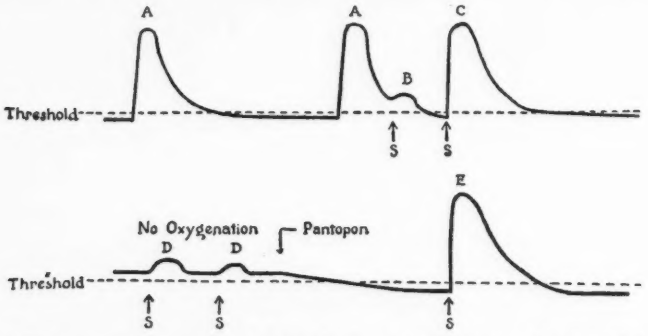

Fig. 1. Diagrammatic representation of relation between state of relaxation of uterus and the type of contraction observed. *A*, Spontaneous maximal contractions, in oxygenated medium. *B*, Submaximal contraction following electrical stimulus. *C*, Maximal contraction following electrical stimulus. *D*, Submaximal contractions following electrical stimuli, during oxygen lack. *E*, Maximal contraction following electrical stimulus, under influence of pantopon. *S*, Electrical stimuli.

the response of the uterus to stimulation on a kymograph. The cathode wire, conducted through an insulating glass tubing, acted as an anchor for the uterine end of the horn at the bottom of the receptacle.

RESULTS

In agreement with the work of O. G. Harne (8) the present results show the existence of two physiologically distinct types of uterine contraction. As Harne has shown, the 'maximal' contractions (large contractions with sudden, steep rise) occur throughout all stages of the estrus cycle, but are somewhat more frequent during estrus. The 'submaximal' contractions are small, or even absent during di-estrus, and characterized by a slow and gradual rise. As estrus supervenes, the submaximal contractions become larger and more frequent until they resemble the maximal contractions in appearance. Following estrus they gradually subside.

The response of the uterus to electrical stimulation apparently depends upon its tonus, as diagrammatically illustrated in figure 1. Above a certain threshold of relaxation the tissue is refractory to the excitation of maximal contractions, and only submaximal contractions may be elicited. This is true whether the stimuli are administered during the course of a previous maximal contraction, or when the base-line remains continually above the threshold as a result of a decreased oxygen supply to the uterus. When it is desired to study responses to electrical stimulation without interference, the latter procedure is used to diminish or eliminate spontaneous activity of the uterus.

Below the threshold, the uterus responds to electrical stimulation with maximal contractions. The base-line of the contractions may be kept continually below the threshold by the addition of an adequate amount of pantopon to the Tyrode's solution. Under these circumstances the uterine responses are exclusively maximal in type. We have ample evidence to show that neither the supply of oxygen nor the exhibition of the above drugs influences the chronaxie of the uterus.

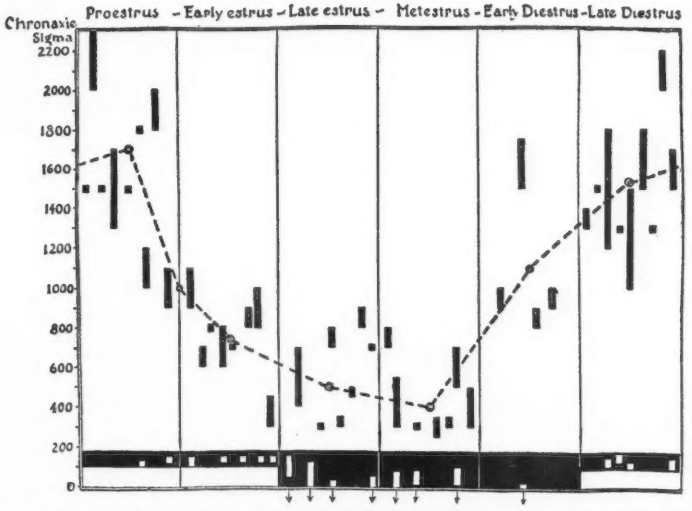

Fig. 2. Chronaxie of the rat's uterus during the various stages of the estrus cycle. Each of the indicated stages occupies approximately 1 day of the cycle. The vertical bars give the chronaxie values, each bar represents a separate experiment, and the length of each bar indicates the range of chronaxie values obtained throughout the whole experiment. The dark bars on the light background represent chronaxie values for submaximal contractions. The broken line running through these bars indicates the average values for all the chronaxie determinations in each stage of the estrus cycle. The light bars on the dark background represent chronaxie values for maximal contractions. The vertical arrows below the maximal contractions, in the late estrus, metestrus and early di-estrus periods, indicate the decrease in chronaxie observed during these experiments, as described in the text.

The electrical excitability of the uterus differs markedly for the two types of contraction described above. The results are graphically illustrated in figure 2. Each vertical bar represents a separate experiment on a rat uterus at the indicated stage of the estrus cycle. The length of the bar indicates the range of values obtained for the chronaxie during that experiment. The dark bars on the white background are chronaxie values for submaximal contractions. The broken horizontal line running through these bars represents the average values for all experiments during each stage of the estrus cycle. The white bars on the dark background are chronaxie values for maximal contractions. There is relatively so little variation in these figures that no averages are given.

It may be seen that the chronaxie of the submaximal contractions varies significantly with the estrus cycle. From an average value of about 1,600 σ during pro-estrus, the chronaxie steadily decreases (increasing excitability) to about 700 σ in early estrus, 500 σ in late estrus, and 400 in metestrus. Thereafter it again increases (decreasing excitability) to reach its previous high

values in the di-estrus. The maximal contractions, on the other hand, maintain a fairly constant, shorter chronaxie (125 to 150 σ) throughout the estrus cycle. But these contractions show another phenomenon during certain phases of the estrus cycle, as indicated by the vertical arrows at the bottom of figure 2. During late estrus and the metestrus, and sometimes in early di-estrus, the chronaxie of the maximal contractions is found to lie in the usual range of about 125 to 150 σ at the beginning of the experiment. But about 3 to 5 hours later the chronaxie begins to decrease, and may almost reach zero after about 5 to 10 hours, when it shows a terminal increase just before excitability is completely lost. (Thus, the apparent low values for some of the experiments graphed for these periods has no real significance, but depend upon how long after excision of the uterus the first determinations were made.) This phenomenon does not occur during the other phases of the estrus cycle, where the chronaxie remains at its initially determined value as long as the uterus remains viable.

DISCUSSION

It is clear that measurements of uterine excitability can be significant only when care it taken to distinguish between the two types of uterine contraction. When this is done, the chronaxie of the submaximal contractions seems to be an index of the physiological state of the uterine muscle, which varies characteristically with the estrus cycle and is susceptible of exact determination.

Chronaxie measurements of the maximal uterine contractions seem to be of less significance, since they do not vary with the estrus cycle. However, the phenomenon of decreasing chronaxie which we have described may be of some interest, since it occurs only during certain stages of the estrus cycle and not at others. The fact that this phenomenon occurs after the uterus has been isolated for many hours suggests that it is of pharmacologic rather than physiologic significance.

SUMMARY

A method and apparatus for measuring the electrical excitability of the isolated uterus, in terms of chronaxie, have been described. The existence of two different types of uterine contractions has been confirmed, and the chronaxie of each determined throughout the normal estrus cycle of the rat. The submaximal contractions show the longest chronaxie (lowest excitability) during di-estrus and pro-estrus, the shortest chronaxie (highest excitability) during estrus and metestrus. The maximal contractions show a constant chronaxie, throughout the estrus cycle, but exhibit a characteristic decrease during experiments conducted in the late estrus and metestrus.

I wish to thank Dr. Samuel Soskin for his valuable aid and advice.

REFERENCES

1. Reynolds, S. R. M.: Physiol. Rev. **17**:304. 1937.
2. Ruebsamen, W., and J. Danziger: Ztschr. f. Geburtsh. u. Gynäk. **69**:656. 1911.
3. Colombo, C., and P. Rowinski: Boll. d. Soc. ital. di biol. sper. **9**:9. 1934.
4. Lapicque, L.: L'Excitabilité en fonction du temps, Presses Universitaires de France, Paris, 1926.
5. Bourguignon, G.: La Chronaxie Chez L'Homme, Masson, Paris, 1923.
6. Evans, C. L.: Excitation and Chronaxie; Recent Advances in Physiology, P. 280, Blakiston, Philadelphia, 1930.
7. Quincke, G., and J. Stein: Ergebn. d. Physiol. **34**:907. 1932.
8. Harne, O. G.: Am. J. Physiol. **99**:227. 1931.

THE POTENCY OF CERTAIN COMMERCIAL HORMONE PREPARATIONS[1]

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During the past few years we have made a study of assay methods for estrogenic, gonadotropic and adrenal cortex hormones, and as part of this study have assayed a number of commercial hormone preparations. It seems desirable to bring together in one paper the results obtained on these preparations, as well as others not reported previously.

It is not the purpose of this report to cast aspersions on the products of any company. As compared with epinephrin or insulin, these hormones are of relatively recent development, uniform assay methods have not been adopted, nor has the problem of deterioration been sufficiently studied. However, we believe that many times the failure of experimental workers to check each other's results and, what is more serious, the failure of one clinician to achieve the same therapeutic success which another has reported, is due to the fact that commercial preparations vary greatly in actual biologic potency. It is to emphasize this fact that the present report is made.

The numerous questions concerning methods and technics are discussed in other papers to which reference will be made. Here, only as much as is necessary for the interpretation of results will be included. All materials were purchased across the counter (as the clinician would purchase them) from the Carey Drug Company and the Imperial Pharmacy, two of the largest prescription pharmacies of Denver.

ESTROGENIC SUBSTANCES

1. Hyroxy-ketonic-estrin. For this form of estrin, the Health Organization of the League of Nations has adopted an 'international standard,' which consists of pooled material from a number of laboratories. This organization has established 0.1 γ of this material as the 'international unit.' The commercial preparations were assayed directly against the international standard material, the same method of assay being used for both. The results are given in table 1.

Comments. The details of the method employed are reported in another paper (1). The oil preparations contained the labelled content, and minor differences between them are probably of no significance. The biologic activity of the aqueous preparations, however, per I.U. stated by the label to be present, was only from one-sixth to one-seventh as great as the biologic activity per I.U. of the international standard material.

[1] This investigation was supported, in part, by a grant from the National Research Council, Committee on Problems of Health.

2. *Tri-hydroxy-estrin.* Only one preparation was studied, theelol, Parke Davis, lot no. 3128030. This was in capsule form and labelled to contain 2000 R.U. or 0.12 mg. of theelol per capsule. The method of assay was not stated. The results of our assay are given in table 2.

Comments. It is generally recognized that the amount of theelol necessary to produce estrus in the castrate rat is very much greater than that required to

TABLE 1. ASSAYS OF HYDROXY-KETONIC-ESTRIN.

Material	Solvent	Method of Injection	Number of I.U. equivalent to 1 R.U.[1]
International Standard	Oil	Single, subcutaneous	13
International Standard	Aqueous	Single, subcutaneous	38
Amniotin (Squibb) Lot No. 54835	Oil	Single, subcutaneous	15
Theelin (Parke, Davis) Lot No. 3053249	Oil	Single, subcutaneous	11
Progynon B (Schering) Lot No. 5-D-18	Oil	Single, subcutaneous	8.6
Theelin (Parke, Davis) Lot No. 3069961	Aqueous	Single, subcutaneous	240.0
Progynon (Schering) Lot No. 5-H-06	Aqueous	Single, subcutaneous	260.0

[1] One rat unit equals the amount necessary to produce an estrus smear in 50 per cent of a large group (25-50) of castrate rats.

TABLE 2. THEELOL—3128030.

No. of Rats	Dosage in Labelled Units	Method of Administration	% in Estrus
	Vaginal Smear Method		
10	100	Subcutaneous	0
10	1000	Subcutaneous	0
10	2000	Subcutaneous	0
10	100	Oral	0
10	200	Oral	40
10	2000	Oral	90
	Vaginal Canalization Method		
9	1	Subcutaneous	33
10	20	Subcutaneous	100

bring about canalization of the vagina in an immature animal. In a recent assay reported by Meyer, Miller and Cartland (2) this ratio was roughly 400 or 800 to 1, depending upon the menstruum. The preparation described above contains approximately the labelled potency on the basis of the canalization method of assay. However, this unit, as shown above, is a very small one as compared to the more familiar estrus smear method. The clinician expecting the same response from 2000 'canalization' units as from 2000 'estrus smear' units would be disappointed. We think it desirable that manufacturers should state the number of units present, according to both methods of assay.

GONADOTROPIC SUBSTANCES

No standard method for the assay of gonadotropic substances has been adopted. We therefore used the manufacturer's method, as described in the literature accompanying the package or on the package itself. Ten commercial preparations were assayed, the results being given in table 3.

TABLE 3. ASSAYS OF COMMERCIAL GONADOTROPIC SUBSTANCES.

Preparation	Source	Assay used by Maker	Our Assay		
			No. Rats	Dosage	% Positive
Parke, Davis, Antuitrin-S					
Lot No. 3115263	P.U.	Lutein'n in 30-day rats	10	1 R.U.	40
Lot No. 3112065	P.U.	Lutein'n in 30-day rats	10	1 R.U.	20
Squibb, Follutein					
Lot No. 56439	P.U.	Lutein'n in 30-day rats	10	1 R.U.	70
Winthrop, Antophysin					
Lot No. 7414	P.U.	Estrus smear, uterine hypertrophy, large foll's and Corp. Lut. in 4 to 5-week-old rats	10	1 R.U.	0
			10	5 R.U.	80
			8	10 R.U.	100
Ayerst, McKenna & Harrison, A.P.L.					
Lot No. R 3880	Plac.	Estrus Smear	10	1 R.U.	100
Searle, Gynantrin					
Lot No. 22633	Pit'y.	Lutein'n in 30-day rats	10	10 R.U.	50
			4	25 R.U.	0
Lot No. 121661	Pit'y.	Lutein'n in 30-day rats	10	10 R.U.	0
Chappel, Prephysin					
Lot No. 101	Pit'y.	50–100% increase in weight of ovaries of 22-day rats	10	2 R.U.	0
			10	4 R.U.	0
Lot No. 626	Pit'y.	50–100% increase in weight of ovaries of 22-day rats	5	10 R.U.	0
			3	25 R.U.	0
Parke, Davis, Antuitrin					
Lot No. 3099247	Pit'y.	No claims made	6	2 cc.	0

Comments. We have carried out an extensive study of the advantages and disadvantages of various methods of assay as applied to substances originating from pregnancy urine, placentae and the pituitary itself (3). From this study it is possible to state the comparative activities which a given preparation shows when assayed by different methods. In the light of this study, the following conclusions concerning the commercial substances may be drawn. The urinary and placental preparations contained approximately the labelled number of units, according to the method of assay employed. The pituitary products, on the other hand, contained relatively little or no activity. However, in rating these activities, the method of assay must be taken into account. It requires less material to produce a given reaction in a 30-day-old animal than in a 22-day-old animal. It requires less material to produce an estrus smear than to produce luteinization. In the case of the placental preparation, the 'unit' represents the *daily* dose, for the others, the total dose. It is therefore obvious that while the labelled number of units may be present, the actual amount of

biologically active substance may vary greatly, depending on whether the unit chosen be large or small. Until the makers of these preparations agree on a uniform method of assay, this unfortunate confusion will persist.

ADRENAL CORTEX PREPARATIONS

The activity of 10 commercial preparations was studied, using the rat as the test animal. The results are given in table 4.

Comments. The commercial preparations reported here were tested in connection with a study of the suitability of the rat for the assay of cortin

TABLE 4. ASSAYS OF COMMERCIAL CORTIN PREPARATIONS.

Preparation	Manufacturer's Assay	Our Assay (Rat)
Wilson, 153	1 cc. equals at least 15 dog units	2 R.U./cc.
Wilson, 156-A	1 cc. equals at least 15 dog units	2 R.U./cc.
Eschatin (Parke, Davis)		
Lot No. 3115857	1 cc. equals at least 10 dog units	8 R.U./cc.
Lot No. 3129959	1 cc. equals at least 10 dog units	Less than 4 R.U./cc.
Lot No. 3144323	1 cc. equals at least 25 dog units	1.6 R.U./cc.
Lot No. 3148364	1 cc. equals at least 25 dog units	2.0 R.U./cc.
Armour, Lot No. 602028	1 cc. contains 2.5 gm. cortex	No activity in 2 cc. daily
Armour, Lot No. 777006	1 capsule contains 2 grains cortex	No activity in 2 caps. daily
Schieffelin, Glyadrenal Lot No. 670670	1 pill contains 2 grains adrenal	No activity in 2 pills daily
Schieffelin, Glycortal Lot No. 670921	1 pill contains 2 grains cortex	No activity in 2 pills daily

(4). We believe individual variations in rats are certain to be less than in dogs, and that the numbers available, ease of handling, etc., make the rat a more desirable animal for this work. A direct comparison carried out by Cartland and Kuizenga (5) shows the ratio of rat unit to dog unit to vary between 1:17.8 and 1:25.4. (Our own studies on experimental preparations agree better with the smaller ratio.) On this basis, it will be seen that the Wilson and Parke Davis preparations were potent, and contained the labelled activity. It should be stated that no claims are made for the activity of the Armour product, nor is it recommended by the makers for use in Addison's disease. An experimental product from this company recently tested by the author showed a high degree of activity. No activity could be demonstrated in the preparations intended for oral administration in the amounts which the rats could be made to eat, although oral administration of a potent product is very effective.

GROWTH HORMONE PREPARATIONS

Four products, from two manufacturer's were assayed using the methods described by them in the circular accompanying the package. Figures 1 and 2 indicate the results.

Comments. It is concluded that these preparations contained considerably less than the indicated potency.

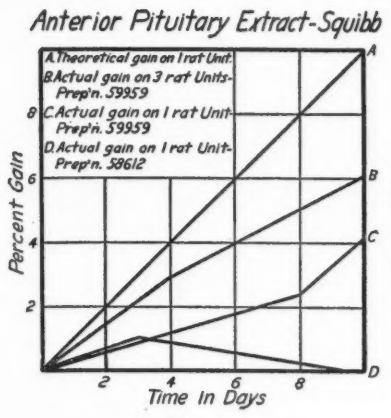
Anterior Pituitary Extract-Squibb
A. Theoretical gain on 1 rat Unit.
B. Actual gain on 3 rat Units-Prep'n. 59959
C. Actual gain on 1 rat Unit-Prep'n. 59959
D. Actual gain on 1 rat Unit-Prep'n. 58612
Percent Gain
Time in Days

Fig. 1.

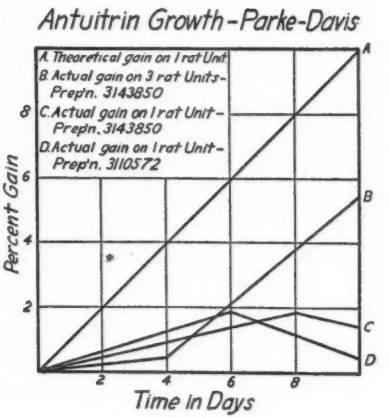
Antuitrin Growth-Parke-Davis
A. Theoretical gain on 1 rat Unit
B. Actual gain on 3 rat Units-Prep'n. 3143850
C. Actual gain on 1 rat Unit-Prep'n. 3143850
D. Actual gain on 1 rat Unit-Prep'n. 3110572
Percent Gain
Time in Days

Fig. 2.

DISCUSSION

The authors believe that the manufacturers of commercial hormone preparations are making an honest and sincere effort to produce dependable therapeutic agents. No one who is acquainted with the men connected with these organizations or who is familiar with the large amount of research which they have devoted to this problem could believe otherwise. At the same time, they themselves would no doubt be the last to assert that complete success had been achieved.

In the light of the present study, we believe that two general criticisms are justified. Extracts of the anterior lobe, both gonadotropic and growth, and aqueous estrin preparations, appear to deteriorate on standing. This problem deserves study and if the time during which activity is retained can be determined it would be well to place an expiration date on the containers.

In the second place, the criticism of lack of uniformity in assay methods and units should be met. In the case of estrin, the Health Organization of the League of Nations has adopted a unit and, in time, official action regarding other hormones will probably be taken. In the meantime, however, to the noncommercial individual, at least, there appears to be no valid reason why the leading firms should not agree upon uniform methods of assay and upon standard units. Other industries have adopted rules of standard practice to which all members conform, with benefit to themselves and to the consumer.

REFERENCES

1. D'Amour, F. E., and R. G. Gustavson: J. Pharmacol. & Exper. Therap. **57**:472. 1936.
2. Meyer, R. K., L. C. Miller and G. F. Cartland: J. Biol. Chem. **112**:597. 1936.
3. D'Amour, M. C., and F. E. D'Amour: J. Pharmacol. & Exper. Therap. In press.
4. D'Amour, F. E., and D. Funk: J. Pharmacol. & Exper. Therap. In press.
5. Cartland, G. F., and M. H. Kuizenga: Am. J. Physiol. **117**:678. 1936.

GONADOTROPIC POTENCY OF THE HYPOPHYSIS IN A WILD MALE RODENT WITH ANNUAL RUT[1]

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Does the hypophysis of male mammals with an annual sexual cycle have the capacity to exert any influence on the reproductive system except during rut? Previous assays with immature female rats as test animals have shown that the hypophysis of the male ground squirrel (Citellus tridecemlineatus) has great sex-stimulating potency during the breeding season, and suggested that it has little or no gonadotropic potency during the non-breeding period (1, 2). These assays were limited both in frequency and in decisiveness by the fact that 6 hypophyses were administered to each test rat.

It was later found that a single hypophysis from a ground squirrel with spermatozoa would greatly stimulate the reproductive organs of an immature female mouse; therefore, this question has been re-investigated, using the mouse as test animal. Hypophyses of 71 male ground squirrels were tested at approximately monthly intervals during 2 annual sexual cycles, and the results are reported in this paper.[2]

MATERIALS AND METHODS

The method employed was essentially that of Clark (3). The hypophysis of a single ground squirrel was cut into fine pieces by scissors, suspended in saline and injected intramuscularly into the leg of a female mouse 21 days of age. The mouse was killed 72 hours later, along with untreated sisters, and weighed. The vaginal membrane was observed for rupture, and a vaginal smear was studied microscopically. Ovaries, fallopian tubes and the uterus of each mouse were trimmed and weighed together while fresh. In 27 treated and 12 untreated mice, serial sections of both ovaries and sections of the uterus were stained and studied.

Table 1 summarizes the number and character of the animals used in this study. For all ground squirrels except some of the bilateral castrates, the reproductive tract of the treated mouse was compared with that of 1 to 5 untreated litter mates, and in this case mice with similar body weight were used. The age of the animals was determined at collection by the use of a number of criteria, most dependable of which was the condition of the prepuce. The prepuce of prepuberal males of this species binds the glans penis so tightly

[1] Aided by a grant from the National Research Council, Committee for Research in Problems of Sex.
[2] The writer is indebted to Dr. Carl R. Moore and Dr. W. C. Allee of the Department of Zoology at the University of Chicago. The former supplied research facilities for some of the assays which were made in Chicago during the summer of 1935, whereas the latter furnished the continuous use of equipment and constant-temperature rooms at Whitman Laboratory.

that the glans cannot be fully exposed by manual pressure, whereas that of adult males at any stage of the sexual cycle permits the glans to be easily exposed.

RESULTS

The character of the annual cycle in males of this species, described elsewhere (1, 2, 4), may be summarized as follows. Males fresh from the field are

TABLE 1. ANIMALS EMPLOYED IN ASSAYS.

Male ground squirrels		Immature female mice	
Young			
Prepuberal	9	Hypophysis recipients	71
With spermatozoa	12	Untreated controls	52
Adult			
Aspermatic	13	Total	123
With spermatozoa	13		
Bilateral castrates	12		
From cold room	12		
Total	71		

in a breeding condition only during April-May, spermatozoa disappearing by June-July and reappearing in March. Laboratory animals produce spermatozoa by December (adult) or January-February (young), whereas those confined in outdoor pens show spermatozoa at approximately the same time as field animals, i.e., March. The presence of spermatozoa is not synonymous with breeding capacity, since the full breeding state is reached about one month following the appearance of spermatozoa. Breeding males kept continuously at approximately 4°C. fail to undergo sexual regression (5).

Controls. Neither weight of reproductive organs nor body weight in control mice was constant; therefore, both should be considered in evaluating the reliability of control data. Weight of the reproductive tract expressed as

TABLE 2. RELATIVE PERCENTAGE WEIGHT OF REPRODUCTIVE TRACT ON BODY WEIGHT IN UNTREATED LITTER MATES.

Litter	Relative percentage wt.	Litter	Relative percentage wt.
1	1.00	8	1.18
2	1.00	9	1.20
3	1.03	10	1.23
4	1.05	11	1.25
5	1.06	12	1.30
6	1.10	13	1.36
7	1.18		
Average for 13 litters	1.15		

percentage of body weight showed great variation for untreated animals of different litters. However, variation between two control animals in the same litter was reasonably small, as is shown by the 13 litters having two or more untreated mice (table 2). The figures in table 2 representing the relative

percentage weight of the reproductive tract on body weight were secured mathematically by the following formula,

$$\left(\frac{\text{Wt. reproductive tract of mouse } 1}{\text{Body weight of mouse } 1}\right) \div \left(\frac{\text{Wt. reproductive tract of mouse } 2}{\text{Body weight of mouse } 2}\right)$$

In other assay litters, only one mouse was reserved as a control animal. It is concluded from table 2 that in order to assume unquestioned gonadotropic potency on the part of the hypophysis in question, a relative percentage weight of the reproductive tract on body weight would have to be greater than 1.36 in a treated mouse.

Young ground squirrels. Young males are born approximately June 1. Their hypophyses show either little or no gonatodropic potency during the first 2 months of life, and low potency until about the eighth month (table 3).

TABLE 3. YOUNG MALES.

Month	Ground squirrels			Reproductive tract of mice		
	No.	Testis mg.	Sperm	Weight mg.	Relative wt.[1]	Relative percentage wt. on body wt.[2]
July	4	17.7	0	14.3	1.47	1.39
Aug.	1	26.0	0	17.8	1.06	1.17
Sept.						
Oct.	1	135.0	0	11.2	1.47	1.37
Nov.	1	301.0	0	14.8	1.76	1.50
Dec.						
Jan.	2	325.2	0	51.9	7.71	6.47
	2	997.1	+	74.4	10.62	9.30
Feb.	7	882.7	+	57.9	5.84	5.62
Mar.	3[3]	780.8	+	68.4	4.24	3.81

[1] Absolute weight of reproductive tract of control mouse represents unity.
[2] Percentage weight of reproductive tract on body weight for control mice counted as unity.
[3] Two of 3 kept in outdoor pens.

Vaginal membranes were either thinned or intact in all assays made in July, partially ruptured in assays during August-October, and completely ruptured in all assays made during the November-March period. It is recognized that the assay mouse received less hypophyseal tissue in the case of very young animals than in the case of older prepuberal males. Data in table 3 indicate that two prepuberal males during January, hence at approximately 8 months of age, had hypophyses with great gonadotropic potency. Condition of the reproductive organs in these two males was at a level which indicated that these assays were made 3 to 4 weeks before the date of spermatozoa appearance, and about 6 to 8 weeks before the date of expectance for full breeding capacity. Hypophyses of all young males with spermatozoa stimulated significantly the reproductive tract of the recipient mouse, the least stimulation being shown for two ground squirrels kept in outdoor pens.

Adult ground squirrels. Adult males are those known to be more than 10 months of age at the time of collection, and they either possessed spermatozoa on the date of collection or had produced them previous to that time.

Adults with spermatozoa. The hypophysis of each of the 13 males in this category stimulated significantly the reproductive tract of the recipient (table

4). However, the relative weight of the reproductive tract on body weight varied from 2.22 to 10.58 for individual recipients. It seems highly probable that this was due somewhat to a difference in sensitivity on the part of different mouse litters, since hypophyses of two ground squirrels, judged by many criteria as being at a similar reproductive level, stimulated the mouse tract to divergent degrees. All assay animals except one had ruptured vaginal membranes. One recipient exhibited a copulation plug and spermatozoa in the vagina, from coitus with her father.

Aspermatic adults. Table 4 shows that the reproductive tract of mice

TABLE 4. ADULT MALES.

Month	Ground squirrels			Reproductive tract of mice		
	No.	Testis mg.	Sperm	Weight mg.	Relative wt.[1]	Relative percentage wt. on body wt.[2]
Apr.	1	1045.0	+	50.0	7.14	7.14
May	4	638.6	+	47.7	5.92	5.80
June	1	69.0	0	15.2	1.94	2.17
July	8	115.4	0[3]	34.6	2.80	2.68
Aug.	3	92.0	0	22.1	1.41	1.39
Sept.						
Oct.						
Nov.	1	299.6	0	11.6	1.38	1.17
Dec.	1	294.0	0	17.4	2.63	2.18
	1	585.0	+	67.7	7.96	8.55
Jan.	2[4]	938.5	+	52.0	7.30	5.28
Feb.	1	568.4	+	98.7	9.05	8.61
Mar.	3	789.8	+	95.3	6.98	6.73
Aug.	12	Bilateral Castrates		72.1	4.90	5.07

[1] Absolute weight of reproductive tract from control mouse calculated as unity.
[2] Control percentage weight of reproductive tract on body weight counted as unity.
[3] Residual spermatozoa in the caudad epididymis in 1 of these 8 animals.
[4] Tiny piece of pituitary of 1 animal lost at implantation.

receiving hypophyses from aspermatic adults weighed less than that of mice given glands from males with spermatozoa. Since the difference between tract weights for recipients and control mice was not a great one, data for individual assays are presented in table 5. The relative percentage weight of the reproductive organs on body weight for a recipient mouse in 8 of 11 assays was greater than 1.36, the figure representing the maximum variability for any two control mice within any single litter (tables 2 and 5).

Microscopical study revealed signs of stimulation in reproductive organs of each of the 8 recipients so examined, and this stimulation was more prominent in uteri than in ovaries. When these organs from both recipients and their litter-mate controls were studied microscopically with the history of the mice deliberately unknown, the uterus was identified correctly as belonging to a recipient mouse in 8 of the 8 cases studied, the ovary being judged correctly in only 5 of the cases. Ocular micrometer measurements of the diameter and the epithelial height for such uteri were made. Uterine diameter of recipients was greater than that for untreated litter mates in each instance, while the height of uterine epithelium in the recipient exceeded that for the

control mouse in only 6 of the 8 cases. The vaginal membrane was fully ruptured in 5 of the 12 assays made during June-November, partly ruptured in 3, thinned but intact in 3 and unmodified in 1.

Bilateral castrates. Twelve males were bilaterally castrated postpuberally for periods of 100 to 235 days and killed during August, i.e., at a time when normal adults show aspermia and complete regression of accessory reproductive organs. The hypophysis of each was assayed, and the results are presented as averages for the entire group in tables 4 and 5. The relative percentage weight

TABLE 5. ASSAYS OF HYPOPHYSES FROM ASPERMATIC ADULTS KILLED DURING JUNE-NOVEMBER.

Date	Testis mg.	Reproductive tract of mouse			
		Weight mg.	Relative weight[1]	Relative percentage wt. on body wt.[2]	Vaginal membrane
June 16	69.0	15.2	1.94	2.17	Thinned
July 12	104.0	59.4	4.72	3.82	Ruptured
" 12	301.0	50.6	4.04	3.58	Ruptured
" 12	77.0	47.6	2.07	2.18	Ruptured
" 15	74.4	15.0	1.23	1.22	Partly ruptured
" 15	100.4	30.4	2.57	2.90	Ruptured
" 20	64.0	25.2	3.07	3.30	Thinned
" 20	66.8	18.6	2.26	2.00	Unmodified
Aug. 7	75.2	19.6	1.16	1.29	Partly ruptured
" 27	110.2	25.2	1.65	1.72	Partly ruptured
" 27	90.6	21.6	1.42	1.18	Ruptured
Nov. 25	299.6	11.6	1.38	1.17	Thinned
		Average	2.29	2.21	
12 Bilateral castrates			4.90	5.07	

[1] Absolute weight of control reproductive tract counted as unity.
[2] Percentage weight of reproductive tract on body weight in control mouse calculated as unity.

of reproductive organs on body weight in individual recipient mice varied from 1.66 to 8.49, only 3 of the 12 assays showing relative values less than 3.00. The vaginal membrane was fully ruptured in 11 of the 12 assays, and unmodified in 1.

Males from cold room. Experimental subjection of males with spermatozoa to a continuous and constant temperature of about 4°C. results in an obliteration of the annual cycle and a persistence of breeding potency (5). Hypophyses of 12 such males were assayed, following periods of 5, 7, 8, 10 and 12 months of treatment. The hypophysis in 11 of these showed much gonadotropic potency, whereas the other exhibited none. In 9 of the 12 assay mice, the vaginal membrane had fully ruptured; and in 2 partially so. The relative percentage weight of the reproductive tract on body weight averaged 5.25 for the entire group of 12 recipients, percentage weight on body weight for untreated control mice being counted as unity in the computations.

Assays for the 3 males killed during August-November, after living continuously in the cold room for 5 to 7 months, are of value because normal adults are in a non-breeding condition during these months. Each of these 3 ground squirrels was anatomically capable of fertile mating, the average weight of one testis being 610.8 mg. The vagina of each recipient mouse was

fully open. The relative percentage weights of reproductive organs on body weight in the recipients were 3.19, 3.22 and 7.78 (average 4.73).

DISCUSSION

Data secured by weighing the reproductive tract from recipient mice and comparing these weights with those of untreated litter mates, body weight also being considered, are not within themselves extensive enough to withstand statistical scrutiny. However, they are supported by microscopical study of ovaries and uterus in crucial cases, and observations on the vaginal membrane of all mice. All of the above data have been considered in evaluating the gonadotropic potency of the hypophysis.

Results of this study demonstrate that gonadotropic potency in hypophyses of prepuberal males was either slight or absent during the first 2 months of life, detectable during August-October, and great at least several weeks before the date of spermatozoa expectancy; in young males with spermatozoa it was also great. Hyphophyses of normal adults, anatomically capable of fertile mating, had great sex-stimulating capacity.

The assays also show that the hypophysis of normal adults has a greatly diminished gonadotropic potency for a long interval of several months during the annual sexual cycle, i.e., the period of aspermia and regressed secondary reproductive organs. Yet slight potency at least was detected in the case of each non-breeding adult. This observation for the hypophysis supports the previous contention that male reproductive organs at the lowest point in the sexual cycle are not in a condition of absolute stasis or complete inactivity (1, 2).

Seasonal diminution of gonadotropic potency was either inhibited or prevented by subjecting breeding males to continuous and constant low temperature. Bilateral castration of breeding males likewise prevented the seasonal diminution in gonadotropic potency. The latter finding agrees with that obtained when immature female rats were used as test animals (2); in the bioassays with rats as test animals it was found that hypophyses of adult males castrated during the non-breeding period also had high gonadotropic potency.

The annual variation in gonadotropic potency is synchronous with the seasonal variation in the cytology of the hypophysis (1, 6), and also with the annual fluctuation within the reproductive organs of the male.

SUMMARY

Hypophyses from 71 male ground squirrels (Citellus) were assayed at approximately monthly intervals during 2 annual sexual cycles. An immature female mouse was given a single hypophysis intramuscularly, killed 72 hours later and weighed. The percentage weight of reproductive organs (ovaries, fallopian tubes and uterus) on body weight for recipients was compared with that of untreated litter mates. In crucial cases, ovaries and uterus were studied microscopically. The vaginal membrane was examined for rupture.

Gonadotropic potency of the hypophysis from adult males showed great seasonal variation, being high in males with spermatozoa and extremely low in adults sacrificed during the long period of aspermia and regressed secondary

reproductive organs. This variation runs parallel to the seasonal cytological fluctuations within the hypophysis which have been previously reported, and also the seasonal variation within male reproductive organs. The fact that the hypophysis showed greatly diminished potency during the long aspermatic period, rather than an absence of potency, supports the previous contention that male reproductive organs are not entirely inactive during the non-rutting period.

Seasonal reduction of gonadotropic potency was prevented, or at least inhibited, both by bilateral castration and by subjection of breeding adults to continuous and constant low temperature.

Potency of the hypophysis from prepuberal males was either slight or absent at 2 months of age, low at 3 to 5 months, and high several weeks before spermatozoa expectancy.

REFERENCES

1. Moore, C. R., G. F. Simmons, L. J. Wells, M. Zalesky and W. O. Nelson: Anat. Rec. **60**:279. 1934.
2. Wells, L. J.: Anat. Rec. **62**:409. 1935.
3. Clark, H. M.: Anat. Rec. **61**:175. 1935.
4. Wells, L. J., and C. R. Moore: Anat. Rec. **66**:181. 1936.
5. Wells, L. J.: Anat. Rec. (Suppl.) **64**:138. 1935.
6. Nelson, W. O.: Anat. Rec. (Suppl.) **64**:34. 1936.

CASE REPORTS

FEMINIZATION IN A MALE ADULT WITH CARCINOMA OF THE ADRENAL CORTEX

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The relationship between the adrenal cortex and sex has been summarized and discussed in previous publications (1, 2, 3, 4). In the adult female, masculinization (virilism) developing with an adrenal cortical tumor is not so very rare, but feminization of the male is sufficiently rare to have its existence doubted. Actually, 5 definite cases of feminization associated with an adrenal cortical carcinoma have been reported, and the present communication adds a sixth. In one case (5) there was a return to normality after removal of the tumor.

Bittorf's (6) case, in 1919, was the first recorded. In August 1918, a male aged 26 noticed enlargement of his breasts. A few months later, there was a decrease in the size of the testes, and a waning of the libido. These changes progressed until February 1919, when he was impotent. In March 1919, a swelling (tumor) was noticed on the left side of the abdomen. Seen by Bittorf in April 1919, the patient was described as well-nourished, although without gross adiposity; the breasts were much enlarged and of a feminine type, with apparently good development of glandular tissue. The external genitals were small. In May 1919, the liver enlarged (metastases), and there was evidence of compression of the inferior vena cava. Cachexia and death followed. At autopsy, Matthias (7) found a 'malignant cortical hypernephroma'. The mammae consisted of loose connective tissue, and true mammary glandular tissue. The testes were extremely small.

The patient in Zum Busch's (8, 9) case was a male aged 27, whose first symptom was painless swelling of both breasts, with development of the nipples, from which some milky fluid could be expressed. One month later, malignant glands were noticed on the left side of the neck, and biopsy indicated that these were secondary to a 'hypernephroma'. Death occurred some 2 months after the onset, and autopsy showed a malignant tumor of the left adrenal cortex, with metastases in the lungs, pleura and mesenteric glands. Sections of the breast tissue gave a histological picture of female secreting glands.

The patient in Holl's (5) first case was a lad of 15 years, in whom the first symptom was said to have been a swelling (tumor) in the right upper abdomen. When seen 2 months later, the breasts were obviously enlarged and projected in bud-like fashion, like those of a young girl. The nipples were

deeply pigmented, and there was also some pigment along the linea alba. The suprapubic hair was of the feminine type, with a sharp horizontal upper border. There was a large tumor in the right loin and upper abdomen. At operation, 2 months after the onset, an irremovable large adrenal tumor was found adherent to the abdominal wall. Material obtained by puncture at operation showed it to be a malignant adrenal cortical tumor. Cachexia and death followed within a few weeks, but there was no autopsy.

The patient in Holl's (5) second case was a male, 44 years old, with 2 children. His sex function was normal until his illness. In the Spring of 1927 both breasts slowly enlarged and became painful. The appearance soon resembled that of female breasts, the nipples becoming large and pigmented, and veins developing over the mammary region. The testes and penis became smaller, libido was lost, and sexual intercourse ceased. The adiposity increased, the breasts became larger and more painful, and the nipples broader and more deeply pigmented. Both the penis and testicles were very small.

On July 16, 1929, a malignant adrenal tumor was removed. A week after the operation, the breasts were less sensitive. One month after the operation a penile erection and pollution occurred; a few months later, libido returned and sexual intercourse was resumed. The breasts became smaller, the adiposity was less, and a varicocele disappeared. In January 1930 (6 months after operation) the patient was well and normal; the penis and testes had grown to normal dimensions, the breasts were small and insensitive, the nipples appeared normal, and their degree of pigment not unusual; and the facies had lost the feminine appearance.

The patient in Lisser's case (29) was a male, aged 33 years, who complained of pain in the left flank and enlargement of the breast, from which a watery secretion could be expressed. Metastases were present in the lungs. Autopsy showed an adrenal cortical carcinoma, apparently arising from a retroperitoneal adrenal 'rest,' since the adrenal on that side was separate from the tumor, and intact. Unfortunately, no mention is made of the state of the genitals or of sexual function.

PRESENT CASE

The patient, *X. Y. C.*, was a physician, aged 34, with one child. Apart from scarlet fever at the age of 10 and quinsy at 29, he was in good health until November 1933, when pain below the left scapula began. This was noticed on waking in the morning, recurred at odd times throughout the day and lasted some minutes or hours, but was unrelated to food or exertion. After a barium meal, radiography showed a small gastric ulcer, and alkaline treatment with diet was given for some months without relief. The pain then radiated forwards along the left costal margin. At this time it would appear that the pain, together with a vague dyspepsia and lassitude, were the presenting features, but on taking the history in retrospect a year later the following points emerged. In 1932 the breasts began to enlarge, and in August 1933, there was loss of libido and partial impotence. Erection of the penis, and coitus, were achieved with difficulty and the genital organs had become smaller. There were also big fluctuations in weight. Whereas the normal weight up to 1932 was 154 lbs., in the next twelve months there was a rapid gain up to 182 lbs., and then a still more rapid fall to 126 lbs. in August 1934.

In July 1934, the patient entered Ruthin Castle, and Sir Edmund Spriggs and Dr. S. W. Patterson, have kindly permitted us to quote extracts of their findings. The patient looked ill; his blood pressure was 116/70, pulse 70. There were no abnormal signs in the chest. His tongue was clean, and his throat was normal. There was some tenderness, on deep pressure, along the left costal margin anteriorly, and the left kidney was low and easily palpable. The spleen appeared somewhat enlarged to percussion, and there was a feeling of resistance (or a mass) above the left kidney, and to its inner side. X-ray examination showed an ill-defined upper pole of the left kidney with a rounded shadow impinging. After a barium meal the stomach was dilated, with evidence of compression just below the cardiac end. Pyelography disclosed normal pelves on both sides; the left kidney appeared to be pushed down, with a rounded shadow superimposed upon its upper outline; the bladder was more like that of a female than a male. Cholescystography revealed numerous large inopaque shadows.

The Wassermann reaction was negative; the sedimentation rate was raised to 14 mm. per hour; blood urea was 39 mg. per cent. A blood examination showed haemoglobin 80 per cent; erythrocytes 4,600,000; leucocytes 7,400; polymorphs 46 per cent, and lymphocytes 49 per cent.

The diagnosis was gallstones, and an ill-defined tumor in the left hypochondrium.

Surgery. In July 1934, one of us (C. A. J.) removed a large malignant tumor of the left adrenal gland. The probability of the tumor being adrenal and malignant made it desirable to select an approach which would facilitate complete extirpation while permitting an examination of the liver and the other abdominal viscera for the presence of metastases. The lumbar route, which is satisfactory for partial or complete unilateral adrenalectomy for hyperplastic conditions of the gland, and which has also been applied to bilateral lesions by Young (10), gives an altogether inadequate exposure for tumors of large size, and renders detection of visceral metastases impossible. The lumbar route with resection of the 12th rib has been used for an adrenal tumor of moderate size, but even in this case the removal of the friable and vascular mass was difficult, and associated with a greater loss of blood than is legitimate.

The transpleural transdiaphragmatic route, which is recommended by some authors, appears to be more hazardous and to give a less satisfactory access to the blood supply of the malignant mass than does the abdominal route. The latter was therefore employed. A left paramedian incision from xiphisternum to umbilicus was combined with an incision at right angles to the former at the subcostal level, and passing backwards to the paravertebral groove. The tumor was exposed without great difficulty after introducing several large gauze packs to displace the coils of the small bowel. The peritoneum over the tumor was incised and the kidney was found to be pushed downwards and overlapped by the neoplasm; but the connections between the two were not intimate, and blunt dissection sufficed to separate them. During this procedure there were only one or two small vessels requiring ligation.

The main vascular connections of the tumor were in relation to its postero-superior surface in close contiguity with the diaphragm. A large curved aneurysm needle was used to control several vascular pedicles, and, following the division of these, hot packs wrung out in saline were firmly pressed into the cavity resulting from the removal of the tumor. Bleeding was well controlled, and the wound then closed in layers with a small drain in the posterior extremity of the transverse element of the incision.

This method of approach to adrenal neoplasms was also adopted by Coller, Field and Durant (11) who also speak well of the access which it affords.

Subsequent progress. Convalescence was somewhat protracted, but after 2 months the patient was well enough to go away to the seashore. He began to gain weight and strength, the breasts became smaller, and the genitals larger. There was some return of sexual urge and potency, but no approach to normality. Courses of deep x-ray were started by Dr. Carter-Braine in October 1934, and continued intermittently. In November 1934, there was acute pain in the left lower chest posteriorly, for one day only. During the next few months, there was gradual improvement in the general condition, and some return of libido.

In March 1935, the pain below the left scapula recurred, and radiated along the costal margin. The patient looked plethoric, the breasts were full, and the genitals rather small. The abdomen was adipose, and lineae distensae, which had appeared early in the illness, were well marked. Blood pressure was 120/80. Sexual desire

TABLE 1.

	Per Liter	
	Comb-growth, Capon units	Estrogens M.U.
Sept. 1934	<50	<100
Nov. 1934	<50	<100
June 1935	<100	>400
Nov. 1935	>40	>400
	>40	>800
	>60	>1600
	>60	>3200
	<100	<6400
Jan. 1936	>25	
	>50	>3200
	<100	<6400

had again declined, and coitus was achieved with difficulty, being followed by partial exhaustion for some days. The hair on the trunk was very profuse and whereas this had always been so the patient thought it had increased with the onset of the illness. On the other hand, the beard growth was, if anything, less strong than previously. (Baldness was a familial feature, and had been present before the illness.) There was no well-defined pigmentation, although the general appearance was dark. Radiation therapy was continued intermittently. The weight continued to increase up to June 1935, when it reached 156 lbs., but from this time onwards there was a progressive fall. Impotence became complete, and the breasts grew larger at first, but then smaller with the general loss of fat. The pain below the left scapula was present intermittently, and in October 1935, the patient felt a sudden pain in the left hypochondrium radiating through to the angle of the scapula. There was a sense of resistance in the left hypochondrium, but no definite tumor.

During the next 6 months there was a progressive decline in the patient's condition, and in April 1936, a laparotomy showed metastatic growth in the liver, which was also clinically enlarged, and a fibrous mass at the original site of the tumor. Dilaudid was given freely, and the patient died in June 1936.

The blood count was estimated several times during the last 2 years of his illness, and although the patient was not taking iron the count persisted at about the following levels: Hb. 80, erythrocytes 4,800,000, leucocytes 7,000. One count in March 1935 showed 5,800,000, and two counts in 1936 showed 3,800,000. It

may be that the secondary anemia, usually associated with malignancy, was hidden by a tendency to polycythaemia which occurs with adrenal cortex tumors.

Biological investigation. Unfortunately, pre-operative studies were not carried out. The day after the operation, Dr. Harold Burrows investigated the estrogenic content of the urine. Six castrated male mice were given 1 cc. of urine morning and evening for 3 days, and on the morning of the fourth day, a total of 7 cc. On the afternoon of that day all gave positive vaginal smears. Eight castrated male mice were given 0.5 cc. doses in similar manner, with positive vaginal smears resulting in all. The test was not continued with smaller doses, and Dr. Burrows concluded

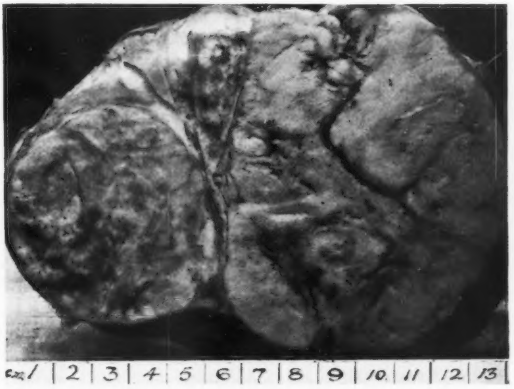

Fig. 1. Malignant adenoma of left adrenal, showing tumor tissue in lumen of vein.

that there was a minimal content of estrin in the urine of 500 M.U. per liter. In view of latter investigations, it seems probable that even a higher content was present.

From September 1934 until January 1936 a series of quantitative estimations of comb growth and estrogenic hormones of the urine were undertaken by Dr. P. de Fremery. The methods used have been described in a previous publication (2), and the results are given in table 1. In September and November of 1934 the amounts of comb-growth and estrogenic hormones were less than normal, or possibly absent. This time the patient was still making a tardy recovery from the operation, and his general condition was improving, although not satisfactory. The figures for June 1935 do not show any gross abnormality, but they are not sufficiently informative. In November 1935 there was a considerable pathological excess of estrogenic hormone (greater than 3000 U. per liter) and a slight increase in comb-growth hormone. Similar data were obtained from the urine in January 1936. Subsequently, all the urine was sent to the Cancer Hospital for attempt at extraction and identification of the estrogenic hormones. This was carried out by Burrows, Cook, Roe and Warren (12). A summary of their findings follows:

"An attempt to isolate the estrogenic hormone from the urine has not been successful, but the results are consistent with the view that the hormone is estrone. From 30 liters of urine were isolated 0.75 gm. of p-cresol, and 3 ketonic compounds which have not hitherto been recognised as urinary constituents. The chief of these, of which 0.4 gm. was isolated, has been shown by analysis and by its artificial preparation by dehydration of dehydroandrosterone to be $\Delta^{3:5}$-androstadiene-17-one. This ketone had weak comb-growth promoting activity in capons, but had

no estrogenic activity or cortical hormone activity. The other 2 compounds were isolated only in amounts insufficient for adequate characterisation, but analysis of their derivatives suggests that they are represented by the formulae $C_{19}H_{28}O_3$ and $C_{21}H_{32}O_3$. Neither of these had appreciable estrogenic activity."

The return of a high content of estrogenic hormone in the urine, which coincided with a decline in the patient's clinical condition, was the first evidence of a return of the malignant growth; clinically, there had been at first no certainty

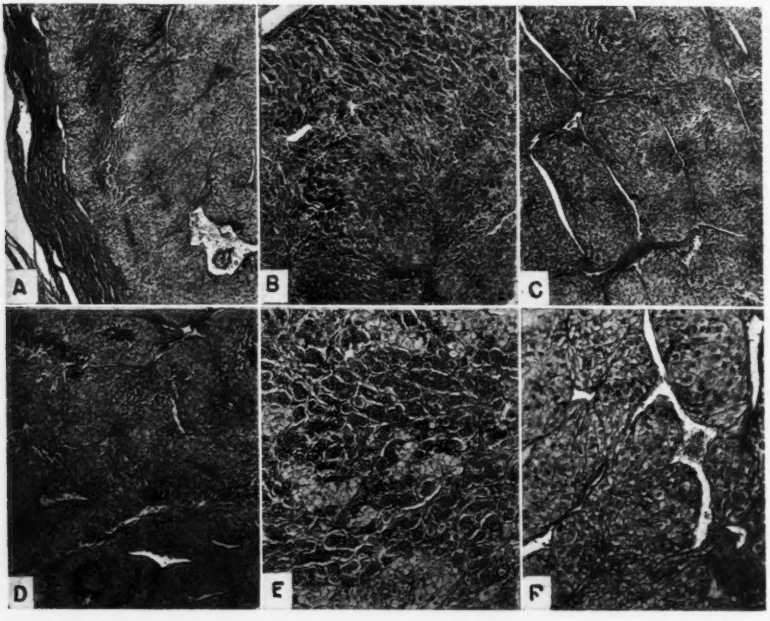

Fig. 2. *A*, Photomicrograph of periphery, showing the well-formed fibrous capsule surrounding the new growth. x50. *B*, Area showing a general resemblance to adrenal cortical tissue. x50. *C*, Another area in which the new growth forms masses of uniform size and shape separated by endothelium-lined blood channels. x50. *D*, A part in which the tumor has a perithelial arrangement. x50. *E*, An area in which groups of 'foam' cells are interspersed among columns of vacuolated cells. x105. *F*, Showing the intimate relation between the endothelial cells of the blood channels and the new growth. x120.

about this. The disappearance of abnormally large amounts of estrogenic hormone from the urine after removal of the tumor, and the reappearance with recurrence of growth, suggests that the tumor was its source.[1] It is interesting that the amount of comb-growth hormone also returned to a urinary concentration slightly above normal at a time when the patient was on the decline. His genitals were then somewhat atrophic, and his libido and potency almost absent. This is possibly explained by the supposition that the adrenal tumor, and not the testes, was the source of the comb-growth hormone.

In June 1935, the Ascheim-Zondek reaction was negative, no trace of ovarian stimulation being observed. In January 1936, it was again negative, but some enlargement of the follicles was noted, although the weight of the ovaries remained within normal limit. It cannot be said that there was any real evidence of follicle stimulating substance.

The pathological report by Miss Leila M. Hawksley, describes "a bilobed tumor,

[1] Callow and Parkes (13) obtained estrin and progestin from the adrenal glands of cattle.

14.5 x 11 x 8 cm., weight 656 gm., with a smooth fibrous outer layer, the lower, smaller incompletely encapsulated mass spherical, 6.5 cm. in diameter, the upper lying transversely. No normal adrenal tissue identified. On section, the tumor is white, soft, and friable, with occasional orange-tinted streaks and areas of haemorrhage, and shows some slight lobulation. At the hilum the large emergent vein, patent at the divided extremity, is largely filled with solid tissue. From a complete slice through one part of the bisected specimen, some 12 or 14 pieces of the tumor tissue were taken for section.

"*Microscopical Examination.* Variations in the structure of the various lobules occur, with differences both in the arrangement and in the staining qualities of the cells. In some parts there is a well-marked resemblance to a normal adrenal cortical structure, with differentiation in all three zones, while through the bulk of the tissue a structure approximating to one or other of the zones is readily recognised. Endothelium-lined blood channels separate the smaller groups and wider columns of tumor cells; the latter, when cut transversely, having an apparently perithelial arrangement. Variations in the staining qualities include a finely granular acidophilic cytoplasm which predominates in the well-vascularized tissue, and a high degree of vacuolation in the larger strands and plaques. No fuchsinophil cells have been detected, an observation which has been confirmed by Dr. H. W. C. Vines. The nuclear characters are fairly uniform, and very few mitotic figures are found. Scattered intra and extra-cellular granules of golden-yellow pigment are present in most fields.

"In all of the sections which include the outer surface the capsule is sharply defined, but in the lumen of the large vein tissue similar to that constituting the main growth is present. The histological features are those of adenoma malignum of adrenal cortical origin, the high degree of differentiation attained suggesting that the tumor is one of a moderately slow rate of growth, and that clinical evidence of the development of the blood-borne metastases may be delayed for some time."

Histological comparison with an adrenal tumor from a patient with virilism. A female patient, aged 34, under the care of Dr. Dorothy C. Hare (14) was found to excrete more than 200 U of comb-growth hormone per liter of urine (2). She died within a few days after removal of an adrenal carcinoma. This event afforded an excellent opportunity to ascertain if there were any histological explanation of the fact that an adrenal tumor in the male appeared to produce an excess of estrogenic hormone, and one in the female an excess of comb-growth hormone. Miss Joan Ross kindly made a comparison of the histological features of the tumors of this and the Hare case of virilism (14).

"In both tumors there are areas in which the structure is that of the classical hypernephroma. The cells in these areas are characteristic, with vacuolated cytoplasm recalling the appearance of the spongiocytes in the outer zone of the cortex of the normal human suprarenal (15). In other areas the two tumors differ.

"In Hare's case areas were present where the tumor consisted of cells arranged in less orderly fashion. The cells varied considerably in size and shape. The cytoplasm was finely granular and stained well with eosin. The nuclei were darkly staining and irregular. Many of the cells showed discrete granules with distinctive staining reactions in the cytoplasm. With Bensley-Cowdray's technic, or Kull's method, the granules were fuchsinophil; with Heidenhain's haematoxylin they stained black and they could also be demonstrated with Mallory's phosphotungstic acid haematoxylin. These granules were found not only in the anaplastic areas of the primary tumor (operation material) but also in the secondary growth obtained post-mortem. Helly Maximow's fixative was used.

"In the present case, variation from the structure of the hypernephroma is towards the formation of a type of cell which is cubical or spheroidal. The cells are arranged in small groups or narrow columns. The cytoplasm of these cells is finely granular and stains deeply with eosin. With Kull's technic or Bensley—Cowdray's stain the cytoplasm is diffusely fuchsinophil and shows no discrete granules.

"While it is evident that these two tumors present a different type of cell there is no definite evidence as to the nature of the difference. Similar granules have been demonstrated (using similar technic) in the cells of the tumor of Lescher's (16) case and another case of adrenal tumor with virilism and the diffusely fuchsinophil cell characteristic of the present tumor is not found in any of these three cases of tumor with virilism.

"It would seem that these two tumors present a different specialized cell type, possibly indicating an origin from, or a tendency to develop into, cells characteristic of different zones of the adrenal cortex (17).

"In a small series of adrenals obtained in the postmortem room and examined by similar technic, differences can be demonstrated between cortical cells of the two sexes. The available material is not sufficient to more than indicate that the differences are fairly constant. In males during, or after puberty, granules are present in the cells of the inner layers of the cortex. In females, granules have been found in groups of cells in the cortex in cases over 50 years of age. No granules have been found in the cortex of younger females, nor do they occur in the adrenals of still-born infants of either sex. This may be a significant fact if substantiated further."

DISCUSSION

The relative frequency of virilism in women compared with feminization in men is usually ascribed to the fact that the ovary is a morphologically bisexual organ, the cortex being feminine; and the medulla potentially masculine. It has also been suggested (3) that the adrenal tumor (hyperplasia or tumor) inhibits the homosexual component, and that the latent heterosexual component then becomes active. Refuting this theory is the fact that virilism may occur after, or even be accelerated by double ovariectomy (18), and likewise the existence of feminization with adrenal tumors, since the testis is morphologically not a potentially bisexual organ. An alternative explanation is that the adrenal cortex itself is more directly responsible for virilism or feminization, a conclusion which was arrived at from a clinical-biological study of virilism (2). It was found that an excess of 'male' comb-growth hormone was found in the urine of 4 of 7 women with an adreno-genital syndrome, 3 women with Cushing's syndrome, and 2 of 3 girls with pseudohemaphroditism. Subsequently, W. J. B. Slot (19) found an excess of comb-growth hormone in the urine of a virile woman with an adrenal carcinoma, but no excess of such hormone in the tumor itself. The latter fact does not exclude the possibility of the tumor giving rise to the hormone since, for example, a resting colloid thyroid contains more thyroglobulin than a hyperplastic one. Gallagher (see appendix to paper of Cahill and others (20) found an excess of comb-growth hormone in two virile women. Callow (21) isolated transdehydroandrosterone from the urine of a virile girl with an adrenal tumor. Kepler (21) found considerable male comb-growth hormone in the urine of a girl of 2 with 'precocious heterologous type of puberty,' but

no prolan or estrin. Broster and Vines (23) and collaborators have isolated a new compound of $C_{21}H_{36}O_3$ from the urine of women showing the adrenogenital syndrome.

The present investigation shows that an excess of estrogenic hormone was present in a male with feminization and an adrenal cortex carcinoma. This excess disappeared after surgical removal of the tumor and the diminution of feminine characteristics, but returned with metastases and the intensification of feminine characteristics. It would appear to provide evidence that the adrenal tumor and its metastases were the source of the estrogenic hormone.

Frank (24), Cahill (20), and others have reported some excess of estrogenic hormone in the urine in virilism. There was no such excess in Slot's (19) or Kepler's (22) case, but in the series of Levy Simpson, de Fremery and Macbeth (2) a slight excess of estrogenic hormone occasionally occurred, although it was by no means a constant feature.

If we restrict further considerations to personal experience, that is, the present case, and that of Dr. D. C. Hare (14; 2, case 8), the problem to be faced is why an adrenal tumor appears to produce a considerable excess of estrogenic hormone in a man with feminization, whereas a similar tumor appears to produce a considerable excess of 'male' comb-growth hormone in a woman with virilism. As far as we are aware, pathologists have not hitherto produced evidence of essential differences in the histological characteristics of adrenal cortical tumors dependent upon the sex of the patient in which they occur. Miss Joan Ross's report does call attention to certain differences in the two cases here discussed, and also to differences between normal adrenal glands in male and female. Whether these differences will prove fundamental and be sustained in a larger series of cases, and whether they prove sufficiently fundamental to furnish an explanation of the biological findings (estrogenic and comb-growth substances) must remain a matter for further consideration. If not, then we should enquire into the possibility of the adrenal gland producing a chemical substance, which can be modified by the testis or ovary into estrogenic or comb-growth hormone respectively, a somewhat surprising mechanism. Certainly there is considerable evidence that, in cases of feminization and virilism, the adrenal cortex is partly if not completely responsible for the excess of estrogenic and comb-growth hormones respectively found in the urine.

The character of the changes that result in the individual from this pathological secretion of sex hormone depends upon *a*), the nature of the hormone, e.g., 'male' comb-growth or estrogenic, and *b*), the fact that the rudiments of heterosexual characteristics are present, and are capable of responding to a biological stimulus. Thus, de Jongh and Mulder (25) produced an increase in the size of the clitoris of female guinea pigs by injecting testicular extract, and Zuckerman (28) observed a similar result in female monkeys following injections of testosterone propionate. Frazier and Mu (26) produced a considerable degree of feminization in male rabbits with estrogenic material: the testes and glans penis became atrophic, the prepuce and urethra came to resemble the female pudenda, the nipples and mammæ became hypertrophied, milk could be expressed, and young rabbits suckled. It is

thought that estrone or testosterone produces atrophy of the gonads in the experimental animal by inhibition of the gonadotropic activity of its own pituitary gland (27).

Incidental to this case is the observation that the prostate was not enlarged, nor was there any difficulty with micturition, a finding which may run counter to the most recent theories on the estrogenic origin of enlarged prostate in man.

SUMMARY

A case of feminization of a male aged 34 is described, together with the partial remission of symptoms following removal of an adrenal carcinoma. Death subsequently followed the recurrence of growth and metastases. The patient excreted a considerable excess of estrogenic hormone (probably estrone), which disappeared after operation and returned with the recurrence of the growth. The histological picture showed certain differences from that of an adrenal carcinoma removed from a virile woman, who secreted an excess of comb-growth hormone. The histological differences may explain why, in the one case, estrogenic hormone and, in the other case, comb-growth hormone were excreted. Alternative explanations are considered. The clinical and biological evidence suggests that the adrenal tumor was directly or indirectly responsible for the excess of estrogenic hormone found in this case of feminization. A similar hypothesis was advanced in a previous paper for the presence of an excess of comb-growth hormone in the urine of virile women.

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REFERENCES

1. Levy Simpson, S., A. Kohn-Speyer and V. Korenchevsky: Lancet **2**:1194. 1933.
2. Levy Simpson, S., P. de Fremery and A. Macbeth: Endocrinology **20**:363. 1936.
3. Levy Simpson, S.: Proc. Roy. Soc. Med. **27**:383. 1934.
4. Levy Simpson, S.: Brit. M. J. **1**:229. 1937.
5. Holl, G.: Deutsche Ztschr. f. Chir. **226**:277. 1930.
6. Bittorf, A.: Klin. Wchnschr. **56**:776. 1919.
7. Matthias, E.: Virchow's Arch. f. path. Anat. **236**:451. 1922.
8. Zum Busch, J. P.: Deutsche med. Wchnschr. **53**:323. 1927.
9. Weber, F.: Brit. J. Dermat. **38**:1. 1926.
10. Young, H. H.: Surg. Gynec. & Obst. **63**:178. 1936.
11. Coller, F. A., H. Field and T. M. Durant: Arch. Surg. **28**:1136. 1934.
12. Burrows, H., J. W. Cook, E. M. F. Roe and F. L. Warren: J. Soc. Chem. Industry **55**:1031. 1936.
13. Callow, R. K., and A. S. Parkes: Proc. Physiol. Soc. **87**:16. 1936.
14. Hare, D. C., J. M. Ross and A. C. Crooke: Lancet **2**:118. 1935.
15. Goormaghtigh, N.: Arch. de Medecine exp. et d'anat. path. **28**:277. 1919.
16. Lescher, P. G., and A. H. T. Robb-Smith: Quart. J. M. **4**:23. 1935.
17. Goormaghtigh, N.: Thése University de Gand, 1922. Quoted in "Special Cytology," Ed., E. Cowdry **2**:910. 1932.
18. Kovacs, F.: Monatschr. f. Geburtsch. u. Gynäk. **91**:65. 1932.
19. Slot, W. J. B.: Acta med. Scandinav. **89**:371. 1936.
20. Cahill, G. F., R. F. Loeb, R. Kurzrok and A. P. Stout: Surg. Gynec. & Obst. **62**:287. 1936.
21. Callow, R. K.: J. Soc. Chem. Industry **55**:1030. 1936.
22. Kepler, E. J.: Personal communication.
23. Broster, L. R., and H. W. C. Vines: Brit. M. J. **1**:662. 1937.
24. Frank, R. T.: Proc. Soc. Exper. Biol. & Med. **31**:1204. 1934.
25. De Jongh, S. E., and J. D. Mulder: Endokrinologie **11**:161. 1932.
26. Frazier, C. N., and J. W. Mu: Proc. Soc. Exper. Biol. & Med. **32**:997. 1935.
27. Meyer, R. K., S. L. Leonard, F. L. Hisaw and S. J. Martin: Edocrinology **16**:655. 1932.
28. Zuckerman, S.: Lancet **2**:676. 1937.
29. Lisser, H.: Endocrinology **20**: 567. 1936.

PITUITARY EMACIATION[1]

(VON BERGMANN)

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In 1914, Simmonds (1) described the syndrome of cachexia hypophyseopriva due to destruction of the anterior lobe of the hypophysis, characterized by progressive cachexia, precocious senility, dental caries, low basal metabolic rate, decrease in size of the vital organs (splanchnomicria) loss of pubic and axillary hair, and loss of libido and sexual function. With few exceptions, the disease is confined to adults and almost invariably progresses to a fatal outcome. Only 5 cases have been reported in children (2, 3) and only one was verified at autopsy. Four of the 5 showed improvement with glandular therapy (one received supplementary roentgen irradiation).

In the past two years several female children have been observed who presented the picture of marked emaciation following a progressive loss of weight, in addition to suppressed or retarded sexual development. The clinical picture resembled the early stages of Simmonds's disease, and yet differed in several respects. It occurred in younger individuals, the severe cachexia was not present and, most important of all, the course was not progressive to a fatal termination but was favorably influenced by endocrine therapy.

As we so often associate the pituitary with obesity, just as we correlate the pancreas with diabetes, it is almost a paradox that we should consider the pituitary in relation to thinness, and because we are so accustomed to thinking of the pituitary in relationship to obesity we may overlook the possibility of this organ in another light.

It is not my purpose to discuss the physiology of the pituitary in relation to metabolism and sexual development, but these pertinent facts are known, namely, that experimentally complete ablation of the anterior lobe results in cachexia and retarded sexual development in the immature (and gonadal atrophy in the mature), whereas injury to the posterior lobe or to the region of the hypothalamus with the pituitary intact results in obesity and disturbed water and salt metabolism without effect upon the gonads. It is possible that in so far as fat, water and salt metabolism are concerned, there may be a physiological relationship between the anterior lobe and the intricate hypothalamic structures.

It is conceivable that a suppression of the anterior lobe may occur as a result of a congenital defect, a toxic influence secondary to the infectious diseases of childhood, or it may be due to a functional exhaustion resulting from the added strain of puberty, or a combination of any of these factors.

[1] Presented as a preliminary report before the Regional Meeting of the American Academy of Pediatrics in Chicago, October 15, 1937.

Von Bergmann (4) was probably the first to realize that there existed clinically a marked difference between thinness (*magerkeit*) and the tendency toward emaciation (*magersucht*). He believed the latter to be a clinical entity which must be differentiated from wasting due to carcinoma, tuberculosis and other chronic diseases. He stressed the fact also that too frequently a diagnosis of anorexia and thinness associated with emotional or psychic changes was made without due consideration to the underlying causes.

Various forms of emaciation have been described by other authors. Graves's disease and Addison's disease have been mentioned by Thannhauser and Grafe (5). The difficulty of attributing emaciation to any one endocrine gland is difficult, and thus many non-committing diagnoses have been made, viz., pluriglandular insufficiency and multiple sclerosis of the endocrine glands (Falta). Many physicians have feared to diagnose the most frequent form, namely *pituitary magersucht* (von Bergmann). However, with our newer information relative to the physiology of the hypophysis, we know that it is the governor of the endocrine machine, and that hypophysectomy or destruction of the hypophysis results in atrophy of the gonads, thyroid and adrenals, clinically exemplified in Simmonds's disease. Thus the emaciation may be due to a disturbance in fat and carbohydrate metabolism resulting from altered secretion of fat metabolism, adrenotropic, diabetogenic, and thyrotropic hormones of the anterior lobe. Depressed or delayed sexual function is secondary to insufficient pituitary gonadotropic hormone. The lowered B.M.R. and hypotension are also probably due to secondary changes in the thyroid and adrenals. Therefore, with considerably less trepidation, we attempt to single out certain instances of emaciation on a pituitary basis, but without positive confirmation because our diagnosis is based upon clinical evidence and response to endocrine therapy.

Recently several cases of so-called Simmonds's disease with recovery (2, 3) have been reported, and the question is raised whether these are true Simmonds's disease or *von Bergmann's magersucht,* in view of von Bergmann's statement that "only those cases should be called Simmonds's cachexia which can be verified anatomically." An analogy may be made to adiposogenital dystrophy which was originally described by Fröhlich as being a severe disorder due to tumor. Now the term is used loosely to describe many cases of adiposity with sexual retardation, which either go on to spontaneous recovery or respond favorably to endocrine therapy.

Von Bergmann (4) has well stated that it is possible to conceive of the anterior pituitary lobe which is anatomically intact but whose internal secretion is inadequate, and it is difficult to explain whether a disturbance of the hormonal and neural regulation as a whole may lead to obesity or to *magersucht.*

In spite of the confusion concerning the etiology and pathogenesis of *magersucht* several authors have agreed that etiologically in certain cases the pituitary is the most important endocrine organ. Von Bergmann (4), Wahlberg (6), Rothman (7), Bickel (8), Dogliotti (9), also Robin (10), and Merklen, Aron, Israel and Jacob (11) have isolated from the group of pathological thinness a pituitary form of emaciation which, in many respects,

resembles the early stages of Simmonds's disease, but is distinguished from it by various clinical characteristics and, above all, by the favorable prognosis and therapeutic response.

SYMPTOMS, DIAGNOSIS AND TREATMENT

The disease almost invariably affects young females, at puberty, during adolescence or following pregnancy. It rarely affects the male. Fourteen of the 15 cases reported by von Bergmann (4) and 13 of the 15 by Bickel (8) were females.

The loss of weight and emaciation start and progress slowly over a long period of time, from a few months to several years. There is associated weakness, fatigue and anemia (the anemia frequently and erroneously given as the cause instead of the effect). There is no mental dullness, but psychic and nervous symptoms are not infrequent. The more severe symptoms and signs of Simmonds's disease, such as falling of hair and nails, marked dental caries, somnolence and cachexia, are not observed. A constant finding is amenorrhea which comes on gradually or, in the younger girl, the menstrual period fails to appear. Robin (10) believes it to be significant that in the older girl amenorrhea develops before the anorexia, which helps to explain the pituitary origin of the disease. Von Bergmann advises watching the tall, thin, asthenic girl with chronic anorexia, psychic disturbances and amenorrhea, just as a vagotonic individual should be watched for hyperthyroidism.

The outstanding physical signs are emaciation, retarded sexual development, delayed secondary sexual characteristics or hypogenitalism, and visceroptosis. Hypotension, a low B.M.R. and low fasting blood sugar are characteristics.

As previously mentioned, the disease is prolonged over months and sometimes years, and not infrequently other symptoms appear, such as anorexia toward certain foods; not infrequently there is increased tolerance for sweets. Not uncommonly the patient complains of vague epigastric pains usually following meals. Gastric analysis varies; in some cases there is hypochlorhydria and in others hyperchlorhydria.

In addition to the low blood pressure, the pulse is frequently slowed, but signs of increased intracranial pressure are absent. The heart may be of the 'drop' or asthenic variety and smaller than normal. Roentgenograms reveal a normal or a small sella, as demonstrated in the 4 cases described by Dogliotti (9). There is no distortion or erosion of the clinoid processes.

The carbohydrate tolerance may be increased; however, von Bergmann describes a particular dextrose tolerance curve in which there is a normal rise (differing from the diabetic) but a retarded fall over the 3-hour period. The response to insulin is more severe and prolonged than in the normal individual, which probably can be explained by a decreased secretion of anterior pituitary insulin antagonizing hormone. Because of the increased sensitivity to insulin, its use is not recommended for treatment. The blood shows a secondary anemia and a retarded sedimentation rate excluding an infectious process.

No pituitary gonadotropic hormone or estrin is demonstrated in the

urine (12), indicating a primary pituitary and secondary gonadal insufficiency. Merklen, Aron, Israel and Jacob (11) describe a simple 'histologic test for hypofunction of the hypophysis in certain cases of emaciation,' which in principle resembles our tests for pituitary gonadotropic hormone in the urine. Their test consisted of the injection of 5 cc. of fresh urine from the patient on 3 consecutive days into a female rabbit 3 weeks of age and weighing 160 to 200 gm. The rabbit was then killed, and the thyroid and ovaries examined histologically. Normally, there is a slight increase in activity of the thyroid and ovaries. In hyperpituitarism there was evidence of an excessive stimulation of the thyroid and ovaries and, in hypopituitarism, no reaction in either organ. They reported on 15 cases of pituitary emaciation, in 14 of which the test showed subnormal reactions.

Diagnosis. The diagnosis of pituitary emaciation should be made only after a careful study, and elimination of all other possible causes for the disease.

Simmonds's disease has a tendency to occur in older individuals, the course is progressively downhill and usually terminates fatally. The emaciation develops into a severe cachexia with muscular atrophy. Fatigue and exhaustion are severe, somnolence develops, the skin becomes dry and wrinkled, the nails fall, and the teeth decay. Premature senility usually develops. Roentgenographic examination may reveal changes in the region of the sella indicative of a tumor. Response to replacement endocrine therapy is temporary.

Addison's disease occurs usually in the adult. In addition to asthenia and hypotension there is a characteristic pigmentation of the skin and buccal mucosa, and severe gastro-intestinal disturbances. Frequently there is evidence of a tuberculous or syphilitic infection.

Careful clinical and laboratory examinations should exclude tuberculosis, malignancy, chronic gastro-intestinal disease, blood dyscrasias, hyperthyroidism and diabetes.

Psychogenic and neurogenic disturbances must be carefully eliminated, in view of the fact that they frequently occur during puberty and adolescence. Psychiatric consultation may be advisable.

Treatment. The principles of treatment consist primarily in the activation of the gonads by the anterior pituitary gonadotropic hormone (gland extract) supplemented by estrogenic substance, which has a favorable influence upon the development and re-establishment of the secondary sex characteristics.

The two cases herein reported responded favorably to anterior pituitary gonadotropic hormone[2] in doses of 1 to 2 cc. hypodermically 3 times weekly, and emmenin, 1 dram, one or two times daily. (In addition, *case 1* received APL hormone (antuitrin-S), at the onset of treatment.) Both preparations were given in 3 to 4-week courses with rest periods of 2 to 6 weeks, depending upon the response to therapy and the ability of the patients to return regularly for treatment. Due to changes of residence, *case 2* was treated by her family and other physicians. At present, two other patients are being treated with APE and estrogenic substance with favorable response.

[2] Maturity Factor (anterior hypophyseal gonadotropic hormone), Ayerst, McKenna and Harrison, supplied through the courtesy of Mr. L. J. Nelsen.

Thyroid extract may be indicated as a metabolic stimulant in spite of weight loss. Plenty of rest and a high caloric diet are valuable adjuncts.

CASE REPORTS

Case 1. S.S., a white female, aged 18 years, was born prematurely, weighing 4 lb. Her developmental history was normal. At 15 years, she began to lose weight gradually, and became less active physically and mentally. She complained of frequent headaches, poor appetite and nervousness. At 16½ years she had not menstruated, and the secondary sex characteristics had not made their appearance. She

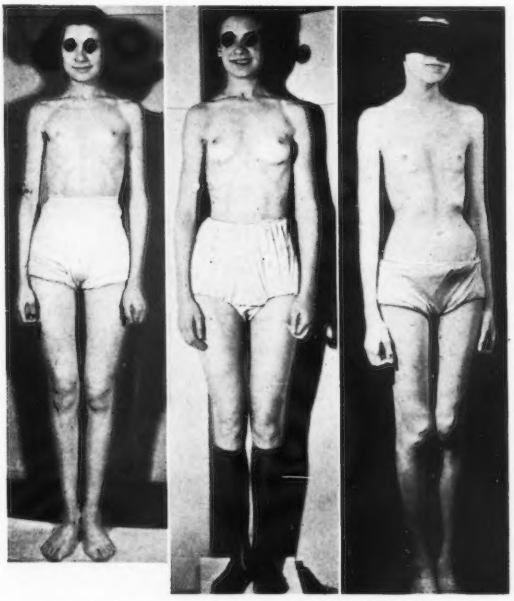

Fig. 1. *Case 1.* 16½ years. Height 57½ inches, weight 69¼ lb. Emaciation, retarded sexual development, delayed menstruation.
Fig. 2. *Case 1.* 18 years. Height 62 inches, weight 97 lb. Growth of 4½ inches and gain of 27¾ lb in 18 months. Normal menstrual periods. Normal sexual development.
Fig. 3. *Case 2.* 18 years. Height 63 inches, weight 81 lb. Emaciation, amenorrhea, atrophy of breasts and genitalia.

tired easily. Because of her immaturity and thinness, she developed an inferiority complex.

Physical examination. The patient at 16½ years measured 57½ inches, weighed 69¼ pounds, and was tall, thin, and emaciated, with a dull facial expression (fig. 1). The breasts were not developed, there was no axillary hair, and the pubic hair was sparse. The external genitalia corresponded to that of a child of 10 years. The thyroid gland was small, the pulse 68, regular, and low tension. The heart was of the asthenic type, a so-called drop heart. Blood pressure was 90/52.

Laboratory findings. Wassermann and Kahn tests were negative, as was the Mantoux test. An x-ray of chest was likewise negative, except for an elongated heart. A roentogram of the skull revealed a somewhat small but otherwise normal sella. The basal metabolic rate was —8, the serum cholesterol 157 mg. Glucose tolerance

was 62, 108, 87, 82, 70 mg. for fasting, ½, 1, 1½, 2 and 3-hr. periods respectively. Quantitative determination of urinary prolan and estrin failed to reveal either hormone. The blood showed a secondary anemia with no eosinophilia.

Course of treatment. From 16½ to 16¾ years of age, the patient received 23 injections of antuitrin-S,[3] with an increase in weight from 69¼ to 73 lb. After 8 injections, she experienced pelvic cramps. From 16¾ to 17 years, she received courses of anterior pituitary gonadotropic hormone (Maturity Factor, Ayerst) 1.0 cc. subcutaneously 3 times weekly. During this period her breasts began to develop, and pubic and axillary hair became more abundant. At 17 years her weight was 77 lb. Between 17 and 17½ years of age she received several courses of anterior pituitary gonadotropic hormone alternated with emmenin orally. At 17½ years she 'spotted' for the first time. Her height was 60 inches, and her weight 81½ lb. At 17 years 7 months, she was given another course of pituitary gonadotropic hormone (Maturity Factor, Ayerst) as she had not menstruated beyond the one period of spotting. At 17 years 8 months, she experienced her first menstrual period. At this time her height was 61 inches, and her weight 86½ lb. During the past 4 months, her menstrual periods have been normal. When last seen, at 18 years, her height was 62 inches and her weight 97 lb. Her secondary sex characteristics were normal (fig. 2), and her mental attitude had improved. Over a period of 1½ years she had grown 4½ inches and gained 27¾ lb.

Case 2. B.P., a white female aged 20 years, a patient of Dr. Julius H. Hess. Birth and developmental history were normal. She was in good health until the age of 15½ years, when she began to lose weight rapidly. In 3 months, her weight dropped from 105 to 60 lb., and she had developed anorexia and vague abdominal distress. As her trouble was thought to be a psychoneurosis because of emotional instability and nervousness, she was removed from school and hospitalized for several weeks during which time, she regained 20 lb. During the past 3½ years, her maximum weight has been 85 lb. She had been hospitalized on several occasions to gain weight, but as soon as she was released she again lost weight rapidly. During her frequent hospital visits, x-ray studies of her chest and gastro-intestinal tract were negative. All other laboratory work was also uninformative. At the age of 16½ years, she menstruated for the first time, just before her weight loss began. Since then she has had scanty periods on two occasions. She has become very depressed over her condition. On various occasions she has worn additional clothing to improve her outward appearance. At 17 years her height was 62 inches and her weight 71 lb., fully clothed.

Physical examination. At 18 years the patient measured 63 inches and weighed 81 lb. (fig. 3). The emaciation was marked, she showed an asthenic habitus, and appeared depressed. The thyroid was small, the teeth in good condition, the chest flat. There was practically no development of mammary tissue; the nipples were small and erect, areola pale pink and small. The lungs were normal, and the heart of the asthenic type, the tones clear, the rate 70, and blood pressure 92/58.

Gynecological examination by Dr. Irving F. Stein revealed sparse hair growth on labia majora and prepuce, but no development of the mons veneris. The clitoris was hypertrophied, erectile and sensitive. The introitus appeared underdeveloped, the hymen was intact. There was slight redness of the external urethral mucous membrane. The labia majora were fairly well developed; the labia minora poorly developed. On rectal examination, the pelvis was easily palpated and free from tenderness. The uterus was very small, probably 3 cm. in length and about 1.5 cm.

[3] 2300 R.U.

across the fundus. The tubes were long, thin and not tender, and the ovaries not definitely palpable. The diagnosis was marked genital underdevelopment suggesting aplasia or extreme hypoplasia of the ovaries and breasts, extremely hypoplastic uterus, and arrested secondary sex characteristics.

Laboratory findings. Wassermann and tuberculin reactions were negative, as were roentgenographic studies of the chest, skull (sella) and gastro-intestinal tract. Gastric analysis (Ewald meal) gave free acid 25, total acid 70. Blood examination showed 4,120,000 red cells, hemoglobin 70 per cent, and white cells 8,350, with a differential of 60 per cent polymorphonuclears, 38 per cent lymphocytes, 2 per cent monocytes. The B.M.R. was —11.6 per cent. Fasting blood sugar was 57 mg. Glucose tolerance was 57, 120, 103, 90, 73 mg. for fasting, ½, 1, 2, 3 hours respectively. Nonprotein nitrogen was 32 mg., cholesterol 218 mg. The electrocardiogram was normal. Examination of the urine for prolan and estrin failed to reveal either hormone.

Course of treatment. After discharge from the hospital, the patient returned to her home in another city. During the past two years she has received periodic treatment with anterior pituitary gonadotropic hormone and estrogenic substance. She has written on several occasions stating that she has gained weight, her appetite has improved and that she feels better. Her menstrual periods became established at 19 years, and are now regular. A letter received from her recently (at the age of 20 years) is very encouraging.

SUMMARY

Two cases resembling the syndrome of pituitary emaciation first described by von Bergmann are reported.

Pituitary emaciation is probably due to hormonal suppression of the intact anterior lobe of the hypophysis, and characterized by its predilection for adolescent girls. The principal symptoms and signs are anorexia and marked loss of weight occurring suddenly or gradually, and prolonged for weeks or months in a girl who has apparently been in good health. Emaciation develops, fatigue and nervousness or psychoneurotic symptoms are frequent, the failure of the menstrual period to appear, or the development of oligomenorrhea are typical. The secondary sex characteristics are delayed, or, in the already sexually mature individual, the breasts and genitalia atrophy. Hypotension, a low B.M.R. and low fasting blood sugar are present. Visceroptosis is usual.

Roentgenograms usually show a normal sella; occasionally the sella appears smaller than normal. The urine shows prolan or estrin in amounts less than normal, indicating a primary pituitary insufficiency. The Wassermann test and the tuberculin tests are negative. There is no evidence of chronic gastro-intestinal disease, or neoplasm. The blood shows only a secondary anemia and a slow sedimentation rate, excluding an infectious process.

The severe symptoms and signs of Simmonds's disease are not present; the course is favorably influenced by endocrine therapy. In our experience, favorable response has occurred with the use of anterior pituitary gonadotropic hormone and estrogenic substance. Thyroid extract may be indicated. Rest and high caloric diet are valuable supportive measures.

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REFERENCES

1. Simmonds, M.: Deutsche med. Wchnschr. **40**:322. 1914.
2. Aldrich, C. A., and J. A. Walsh: J. Pediat. **7**:490. 1935.
3. Dunn, C. W.: J. Nerv. & Mental Dis. **83**:166. 1936.
4. Von Bergmann, G.: Deutsche med. Wchnschr. **4**:123. 1934; *ibid.*, **5**:159. 1934.
5. Thannhauser, G.: Quoted by von Bergmann.
6. Wahlberg, J.: Acta Med. Scand. **84**:550. 1935; J.A.M.A. **106**:1968. 1936.
7. Rothman, H.: Acta med. Scand. **87**:168. 1935.
8. Bickel, G.: Gaz méd de France. **12**:535. 1934.
9. Dogliotti, G. C.: Riv. di clin. med. **37**:302. 1936.
10. Robin, G.: J. d. praticiens. **31**:503. 1936.
11. Merklen, P., M. Aron, L. Israel and A. Jacob: Progrès méd. **48**:1849. 1936.
12. Zondek, B.: Die Hormone des Ovarium und des Hypophysevorderlappen. Berlin, Springer, 1931.
13. Merklen, P., M. Aron, L. Israel and A. Jacob: Bull. et mém. Soc. méd. d. hôp. de Paris. **28**:1360. 1936.

CLINICAL AND LABORATORY NOTES

THE OCCURRENCE OF COITUS-INDUCED OVULATION IN THE ADRENALECTOMIZED RABBIT* †

CHANDLER McC. BROOKS AND SAMUEL W. PAGE

There is much evidence to support the contention that ovulation which occurs in the rabbit following coitus or electrical stimulation of the brain or spinal cord is produced by a substance or substances liberated from the anterior lobe of the pituitary gland.[1, 2, 3] This coitus-induced or stimulation-induced anterior lobe discharge appears to be mediated by nerves passing to the gland from the hypothalamus by way of the pituitary stalk.[4, 5, 6]

The cat, like the rabbit, normally ovulates only on coitus, and it has been assumed generally that a similar mechanism operates in the two species. Recently Friedgood[7] has claimed that in the cat the adrenal glands are involved in the coitus-induced activation of the anterior lobe of the pituitary. The basis for this belief was his observation that removal of the adrenal glands from cats within 15 to 55 minutes after coitus prevented ovulation, but adrenal ablation 6 hours or more after the occurrence of coitus did not abolish the ovulatory response. He concluded that following coitus a cortico-adrenal substance is liberated capable of stimulating or coöperating in the stimulation of the secretion of gonadotropic hormone by the anterior lobe.

We have attempted to ascertain whether or not the rabbit's adrenal cortical tissue plays a rôle in the series of events initiated by coitus and culminating in ovulation. Two types of experimental procedure were followed.

First, 5 female rabbits were mated 2 to 4 times within a period of a few minutes. After being promptly anesthetized, both adrenals were ablated within 15 to 45 minutes after the first mating. This procedure differed from that followed by Friedgood in that one adrenal in his animals had been removed and the animals permitted to recover fully and come into estrus previous to the experimental mating and final adrenalectomy. The one-stage double adrenalectomy was more drastic; nevertheless, all the rabbits ovulated normally, even though they died within 2 to 6 days, presumably from cortical deficiency. In 1 rabbit which died 18 to 20 hours after mating, ova were found in the reproductive tract. In the other animals, eggs were not looked for; the presence of rupture points, corpora lutea and progestational changes in the uteri were considered as adequate proof that ovulation had occurred. Only 1 of the rabbits possessed accessory adrenal tissue. This remnant evidently had not been large enough to sustain life. The fact that these rabbits ovulated indicates either that the adrenal cortex is not essential to ovulation or that sufficient hormone can be liberated within

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† This work was aided by a grant (to Dr. Philip Bard) from the Committee for Research in Problems of Sex, National Research Council.

[1] Fee, A. R., and A. S. Parkes: J. Physiol. **67**:383. 1929.
[2] McPhail, M. K., A. S. Parkes and W. E. White: J. Physiol. **79**:180. 1933.
[3] Marshall, F. H. A., and E. B. Verney: J. Physiol. **86**:327. 1936.
[4] Brooks, C. McC.: Am. J. Physiol. **121**:157. 1938.
[5] Harris, G. W.: Proc. Roy. Soc. London, s. B. **122**:374. 1937.
[6] Westman, W., and D. Jacobsohn: Acta obst. et gynec. Scandinav. **17**:235. 1937.
[7] Friedgood, H. B.: Science **86**:84. 1937.

a 15-minute period following coitus to initiate the required anterior lobe activity.

A second type of experiment was performed which eliminated the possibility of any adrenal activity. Five rabbits were adrenalectomized. Within a few hours after the adrenal removal, the animals were given an intramuscular injection of 20,000 I.U. of progynon-B. In addition to stimulating mating activity under these rather unfavorable conditions, this estrogenic substance probably tended to prevent cortico-adrenal deficiency.[8] Large amounts of sodium chloride were mixed with the food given to these animals. The 5 rabbits were mated several times on the second and third days after adrenalectomy. Two animals died on the fifth day despite the progynon-B and the high salt content of the diet, but 2 others showed only slight signs of cortical deficiency when they were killed on the sixth day after operation. Two of the 5 rabbits possessed small accessory adrenals, but one of these had died on the fifth postoperative day. All of the animals ovulated. Three additional adrenalectomized rabbits were injected with 20,000 I.U. of progynon-B, but were not mated. These did not ovulate. Two normal females, to which an identical dose of estrogenic material had been administered, likewise did not ovulate, indicating that the large quantities of progynon-B injected had not produced the ovulation which occurred in the mated adrenalectomized animals.

These experiments prove beyond question that ovulation can occur in the absence of the adrenal cortex. In chronic adrenalectomized rabbits, changes might occur in the pituitary and reproductive organs which would prevent ovulation on coitus, but evidence is lacking that coitus stimulates any cortico-adrenal activity essential to ovulation.

Summary. Removal of both adrenal glands within 15 to 45 minutes after mating does not prevent ovulation in the rabbit. Completely adrenalectomized rabbits mated on the second or third day after adrenalectomy ovulate normally. The adrenal cortex is not an essential link in the chain of events whereby coitus stimulates the ovulation-producing activity of the anterior lobe of the rabbit's pituitary.

[8] Thorn, G. W., and G. A. Harrop: Science 86:40. 1937.

KERATODERMIAS OF THE HANDS AND FEET AND THYROID DEFICIENCY

JOSÉ M. CERVINO, ALBERTO BERTOLINI AND R. A. LARROSA HELGUERA

The keratodermias, although long known, still present many problems of etiology and treatment. Occasionally the well-known depression of dermal nutrition characteristic of thyroid deficiency shows a regional exaggeration that leads to a keratodermia of the palms and soles. The condition was first described as a clinical entity by Mussio Fournier[1]. We desire to report two additional cases of keratodermia in which thyroid treatment resulted in recovery.

Case 1. L.P., a female aged 15 years, was diagnosed as having congenital hypothyroidism, chiefly evidenced by the following symptoms. Her height was under minimum normal for her age. She had a generalized adiposity, mental retardation, menstrual disturbances characterized by a profuse flow and with long intervals of amenorrhea, sensitiveness to cold, and constipation.

Her skin, thickened throughout, was very dry and harsh, and there was subdermal infiltration, with very intense desquamation. The palms of her hands showed marked hyperkeratosis and chapping. On the soles of her feet the hyperkeratosis was more decided and the chapping deeper. Her B.M.R. was —24 per cent. The Wassermann reaction was negative.

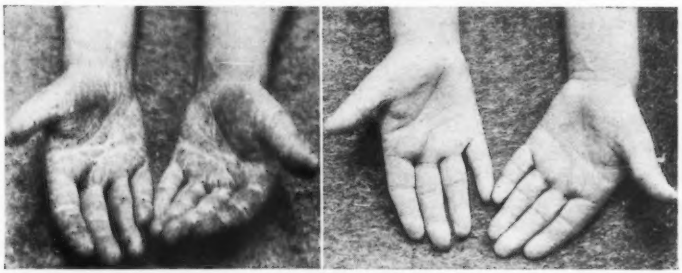

Fig. 1. *L.P., Case 1.* Congenital hypothyroidism, before treatment (11/28/36), and after treatment (1/23/37).

Thyroid substance was given for one month, in amounts varying between 0.10 gm. and 0.20 gm., after which time the keratotic process in the palms and soles disappeared almost entirely.

Case 2. T.P., a female aged 41 years, was diagnosed as having acquired myxedema. The outstanding features were a generalized adiposity, pitting edema about the lower extremities, puffiness, supraclavicular fat pads, sensitiveness to cold, apathy, fatigue, constipation, a hoarse, husky low-pitched voice, metorrhagia, anemia, and a thick dry skin similar to that in ichthyosis. She showed a marked hyperkeratosis of the palms and soles. Her B.M.R. was —30 per cent. The Wassermann reaction was negative, and the electrocardiographic tracing was typical of myxedema.

The patient was placed under thyroid treatment, the dosage gradually being increased to 0.30 gm. daily. Large lambeaux of skin began to fall off the palms and soles, with a concomitant disappearance of ichthyosis and edema. Following this, a remarkable improvement of the entire clinical picture was observed.

[1] Moussio-Fournier, J. C.: Bull. et mém. Soc. méd. d. hôp. de Paris, 48:1236, 1932.

DYOSTOSIS MULTIPLEX

WILLIAM ANTHONY REILLY*

Over a period of years descriptions of similar syndromes have appeared in literature under various titles. These syndromes always involve the skeleton and subcutaneous tissues, and often the eyes, central nervous system and some of the viscera. Children only have been affected. The disease does not seem to be hereditary, but there have been a few instances of familial occurrences.

The skeleton is dwarfed, cortices and periostium thickened and epiphyseal development retarded. Kyphosis and large skulls occur. Extension in the large and small joints is limited. Subcutaneous tissue is abundant, skin thick and 'myxedematoid.' Often there are cloudy corneae, mild optic neuritis, mental retardation and occasionally pathological findings suggestive of amaurosis. Hepatosplenomegaly devoid of characteristic pathological findings of any known disease is also present. Other less constant occurrences are relative normality at birth, early onset at about one year of age, inguinal hernia, and deafness.

The writer has previously reported 3 instances under the title, "An Atypical Familial Endocrinopathy in Males with a Syndrome of other Defects."[1] In this article, reference of similar cases are complete to that date, with 3 exceptions.[2, 3, 4] The variability of titles does not give a searcher of the literature much opportunity to detect these similar reports.

Since my earlier report[1] the title, "Dyostosis Multiplex," has appeared in the American literature in Washington's article.[5] I believe this title should be adopted for the syndrome. Although descriptive of the skeletal findings, it does not cover changes in other parts of the body. Nevertheless, it will serve as a very helpful classification.

Dyostosis multiplex, the hyperplastic form of degenerative dyostoses, also includes: (1) dystrophia periostealis hyperplastica familiaris (Dzierzynski); (2) dysostosis craniofacialis hereditaria (Crouzon); (3) hypertrophic cranniene simple familiale (Klippel-Feilstein); (4) the Helmbolz-Harrington syndrome;[6] (5) Leris' pleonosteosis (as a minor form of skeletal change only). In England, Ellis[8] has described what he terms 'Gargoylism.' This is also dyostosis multiplex.

Some have thought that the syndrome is due to the concomitant occurrence of rickets, hypothyroidism and the mongoloid state. I cannot detect the mongoloid state in the 5 cases which I have followed. The author feels with F. Parkes Weber,[7] of London, that "one might provisionally make another group headed by the type described by Helmholz and Harrington with the author's (Reilly's) cases (dysostosis multiplex) as a less severe variety, and that described by Pritchard as a minor incomplete form." Weber also stated that "he might be able to add some other cases, for instance, some children with enlarged livers and spleens and peculiar facies. I have in mind the cases of Hunter, Sheldon, Ellis, Poynton et al, to which might be added Marriott's cases."[1]

* From the Department of Pediatrics, University of California Medical School, San Francisco.

[1] Reilly, W. A.: Endocrinology **19**:639. 1935.
[2] Hurler, G.: Ztschr. f. Kinderh. **24**:220. 1919.
[3] Binswanger, E., and O. Ullrich: Ztschr. f. Kinderh. **54**:699. 1932.
[4] Pfaundler, M., and A. Schlossmann: Handb. d. Kinderh. **1**:694. 1931.
[5] Washington, J. A.: Brennemann's Practice of Pediatrics, Vol. IV, Chap. XXX, W. F. Prior, Hagerstown, 1937.
[6] Helmholz, H. F., and Ethel R. Harrington: Am. J. Dis. Child. **41**:793. 1931.
[7] Personal communication.
[8] Ellis, R. W. B., W. Sheldon and N. B. Capon: Quart. J. Med. **29**:119. 1936.

ASSOCIATION NOTICE

SCIENTIFIC PROGRAM OF THE TWENTY-SECOND ANNUAL MEETING

Hotel St. Francis, San Francisco, California

The Rôle of the Thyroid in Functional Colon Disorders. Felix Cunha, *San Francisco, Calif.*

Effect of Parathyroidectomy and Renal Function on the Action of Calciferol. F. A. McJunkin, M. C. Patras, R. D. Templeton and W. R. Tweedy, *Chicago, Ill.*

Diabetes Mellitus Associated with Hyperthyroidism. Daniel P. Foster and William L. Lowrie, *Detroit, Mich.*

Insulin Therapy in Mental Diseases. C. J. Kuth and D. V. Conwell, *Halstead, Kans.*

The Effect of Insulin Hypoglycemia on the Central Nervous System. Harold E. Himwich, Karl M. Bowman, Joseph Wortis and Joseph F. Fazekas, *Albany and New York, N.Y.*

Experimental Obesity. Eaton M. Mackay, *San Diego, Calif.*

Pituitary Dwarfism Versus Diabetic Infantilism, Illustrated by a Case Report. James W. Sherrill, *San Diego, Calif.*

A Critical Statistical Comparison of Sixty-nine Pathologically Verified Cases of Simmonds' Disease with One Hundred Thirty-four Clinical Cases. Roberto F. Escamilla, *San Francisco, Calif.*

Infantilism with Congenital Webbed Neck and Cubitus Valgus. Henry H. Turner, *Oklahoma City, Okla.*

Studies Relating to Methods of Characterizing Two Gonadotropic Hormones, Follicle-Stimulator and Luteinizer. F. J. Saunders and E. H. Cole, *Davis, Calif.*

The Relation of the Anterior Pituitary to the Development of the Mammary Glands. Warren O. Nelson, *Detroit, Mich.*

Progestin Studies I. Pregnandiol Excretion. J. P. Pratt and R. F. Stover, *Detroit, Mich.*

The Therapeutic Use of the Sex Sterols in Functional Meno-Metrorrhagia. E. C. Hamblen, *Durham, N.C.*

The Use of Testosterone Propionate in Functional Menorrhagia. Experimental Study of Twenty-five Cases Controlled with Endomentrial Biopsies. Samuel H. Geist, Udall J. Salmon and Joseph A. Gaines, *New York, N.Y.*

The Effect of Androgens on the Genital Tract of Immature Female Rats. Udall J. Salmon, *New York, N.Y.*

Androgens of Blood, Urine and Testes. D. Roy McCullagh and Walton O. Osborn, *Cleveland, Ohio.*

The Rôle of Biochemistry in Endocrinology. E. C. Kendall, *Rochester, Minn.*

The Diabetes of Bearded Women (Diabetes, Hirsutism and Suprarenal Tumor). E. Clare Shepardson and Edward Shapiro, *San Francisco, Calif.*

A Case of Arrhenoblastoma which Simulated Cushing's Disease. C. Kelly Canelo, *San Jose, Calif.*

Pseudo-Sexual Precocity, The Adrenal Cortical Syndrome in Girls. Report of Successfully Operated Case and Review of the Literature. Anthony Reilly, H. Lisser and Frank Hinman, *San Francisco, Calif.*

Subclinical Adrenogenital Syndrome. S. J. Glass, *Los Angeles, Calif.*

Response to Cold Following Double Adrenalectomy. S. M. Horvath, *Oxford, Ohio.*

Adrenal Insufficiency Secondary to Pituitary Tumor. Frank N. Allan, *Boston, Mass.*

The Treatment of Addison's Disease with Adrenal Cortex Extract. W. O. Thompson, *Chicago, Ill.*

Recent Studies in Cushing's Disease. Evelyn Anderson and Webb Haymaker, *Berkeley, Calif.*

Further Studies of Male Sex-Stimulating, Female Sex-Repressive Fractions of the Adrenal Glands of Cows, Steers and Bulls. F. M. Pottenger, Jr., *Monrovia, Calif.*

The Endocrine Aspect of Essential Hypertension and Diabetes Mellitus. James H. Hutton, James T. Case, William L. Culpepper, E. C. Olson and E. E. Madden, *Chicago, Ill.*

Diagnostic Value of Build in Menstrual Disorders and Obesity. Leona M. Bayer, *San Francisco, Calif.*

Endocrine Theories of Dysmenorrhea. C. F. Fluhmann, *San Francisco, Calif.*

Effect of Anterior Pituitary Sex Fraction upon the Development of the Human Uterus. Sheldon A. Payne, E. Kost Shelton and B. N. Tager, *Los Angeles, Calif.*

Observations Associated with Uterine Bleeding. Allan Palmer, *San Francisco, Calif.*

Antigonad Effect of Prolactin in Menorrhagia. George Joyce Hall, *Sacramento, Calif.*

Tuberculosis and the Female Sex Cycle. Emil Bogan, Miriam Hubbell and Josephine West, *Olive View, Calif.*

The Specific Dynamic Action in Castrated and Menopausal Women. William M. Moffat, *Santa Barbara, Calif.*

For further information about the meeting, see Page 386 of the March number.

CURRENT ENDOCRINE LITERATURE

COLLABORATORS

Fuller Albright
Wayne J. Atwell
Geoffrey Bourne
Israel Bram
John C. Burch
H. J. Deuel, Jr.
John C. Donaldson
Maurice H. Friedman
Murray B. Gordon
Arthur Grollman
Frank A. Hartman
E. C. Hamblen
F. C. Koch
Edward Larson
J. T. Lewis
Joseph M. Looney
A. E. Meyer
Warren O. Nelson
J. P. Pratt
A. T. Rasmussen
Elmer L. Sevringhaus
L. B. Shpiner
D. L. Thomson
R. L. Zwemer

ADVANCE ABSTRACTS

Are the lactogenic and carbohydrate metabolism hormones identical? Bergman, A. J., and C. W. Turner. To appear in this Journal.

The separate identity of the lactogenic and the carbohydrate metabolism hormones of the anterior pituitary are indicated by chemical separation and biological assay. A method is described for separation of two fractions, one of which is rich in the lactogenic, and the other in the carbohydrate, metabolism hormone. By biological assay methods, it was shown that the lactogenic fraction contained about 6666 pigeon U of lactogen per gm. by the McShan-Turner method, whereas the carbohydrate metabolism fraction contained only 125 pigeon U per gm. The carbohydrate metabolism fraction contained about 500 guinea-pig U of carbohydrate metabolism hormone per gm. by the Bergman-Turner method, whereas the lactogenic hormone contained only 10 guinea-pig U per gm. The separation and purification of the two fractions described has increased the potency of each extract by about 100% over the assay of the initial extract.

The effect of testosterone propionate on human female castrates. Birnberg, C. H., L. Kurzrok and S. Livingston. To appear in this Journal.

Three patients were treated with testosterone propionate for menopausal symptoms resulting from x-ray and radium castration. Marked improvement was noted in each case. The effect upon the endometrium was studied in two cases in which the previous administration of estrone had been followed by uterine bleeding.

Fatty atrophy following insulin injection in nondiabetic malnutrition. Blotner, H. To appear in this Journal.

This paper presents some observations on local fat atrophy which occurred in a normal, thin person who took insulin for 3 weeks to gain weight. About 4 or 5 months after the insulin was omitted the atrophy was noted in the right thigh, where the drug was injected twice as frequently as in the left thigh. This increased in size and reached its maximum diameter in the subsequent 4 months. During the past 2½ years, the depression in the thigh has remained stationary except during pregnancy, when it appeared more marked. The sensation was diminished over the region of the atrophy. This case illustrates that a patient does not have to be diabetic or fat to develop insulin atrophy, and that the atrophy may appear some months after the withdrawal of insulin. Just why a patient of this type should develop fat atrophy under insulin treatment is conjectural. There is no reason to believe that the cause for it is any different in the nondiabetic than in the diabetic.

Precocious masculine behavior following administration of synthetic male hormone substance. Hamilton, J. B. To appear in this Journal.

In the newly hatched chick, synthetic male sex substance, although causing stunting of body growth, can stimulate not only gross and cytological growth of organs but also, at least

in the males, induction of cock-like behavior. The function of crowing appeared in 10-day old chicks weighing 68-70 gm. and antedated development of mature, polypitch tones. This stimulation of precocious function is of interest in its relation to the influence of androgenic substances upon sexual and other behavior. The data reported are interpreted as evidence that a behavior recognized as 'masculine' can be induced by synthetic male hormone substance and can be accomplished irrespective of the knowledge of an individual that treatment is being given.

Response to cold following double adrenalectomy. Horvath, S. H. To appear in this Journal.

Studies of the respiratory metabolism of normal and completely adrenalectomized Albino rats at environmental temperatures of 29° and 4°C., and at 29°C. after exposure to 4°C. for periods of one to two hours show that the basal heat production of Albino rats is lowered approximately 10% following bilateral adrenalectomy. No stimulation of the heat production measured at 29°C. was observed in adrenalectomized rats exposed to a temperature of 4°C. There was a decrease of 10.6% in comparison to an increase of about 20% in the normals. At an environmental temperature of 4°C., the heat production was increased 79% in the adrenalectomized animals compared to the 176% increase observed in normal rats. Adrenalectomized rats were able to maintain postoperative colonic temperatures for several hours, but failure occurred rapidly thereafter. This fall was due to a decreased heat production.

Adiposity and diabetes mellitus in a monkey with hypothalamic lesions. Ranson, S. W., C. Fisher and W. R. Ingram. To appear in this Journal.

An account is given of a monkey which developed adiposity within 3 weeks after lesions had been placed in the hypothalamus. The weight doubled in 2 months and thereafter increased more slowly. The weight was 2.6 kg. on the day of operation and 9.07 kg. when the animal was killed 17 months later. There was no edema or ascites. At autopsy normal mature testes were found. An associated diabetes mellitus was explained by the finding in the pancreas of hydroptic degeneration of the islands of Langerhans. The lesions in the hypothalamus, and the histology of the hypophysis, suprarenal glands and thyroid were studied without yielding clear evidence concerning the cause of the adiposity. The most striking peculiarity of the brain was the presence of two large bundles of myelinated fibers running from the hypothalamus into the proximal part of the infundibulum, i.e., into the medial eminence.

The effect of castrate urine hormone on the testis. Rubinstein, H. S. To appear in this Journal.

The effect of castrate urine extract (gamone) upon the testis was studied in sexually mature and immature albino rats. Litter-mate brothers served as controls. Test animals received 10 daily (except Sunday) intraperitoneal injections of 10 R.U. of the hormone (total 100 R.U.), whereas controls remained uninjected. Twenty-four hours after the last injection all animals were sacrificed, and body and testicular weights were determined. The testes including epididymes were also studied for microscopic differences. It was found that the hormone used induced premature testicular descent, significantly increased testicular weight, increased the interstitial tissue in both groups, and enlarged tubular diameter in the mature rat. Germinal cell proliferation was increased, but maturity was not hastened.

Cellular changes in the anterior hypophyses of Vitamin A deficient rats. Sutton, T. S., and B. J. Brief. To appear in this Journal.

Differential cell counts were made on anterior hypophyses of normal, Vitamin A deficient, and castrate rats of both sexes, in an attempt to determine the 'modus operandi' of Vitamin A deficiency in causing sterility. It was found that a deficiency of Vitamin A resulted in an increase of the percentage of beta (basophilic) cells. There was also an increase in the number of beta cells containing a macula. This increase was significant in deficient animals of both sexes. However, when compared with normals, a greater relative increase in beta cells was noted in hypophyses from Vitamin A deficient males than from Vitamin A deficient females. The changes in the hypophyses noted in Vitamin A deficiency approach those found in true castrates. The greater relative change in hypophyses of Vitamin A deficient males may be accounted for on the basis of a more nearly complete destruction of the germinal epithelium of the male gonad. Forty-nine rats were used in this study, and a total of 39,269 cells were counted. This study presents further evidence that a Vitamin A deficient diet causes direct damage to the gonad, the increase in beta cells representing a compensatory change in the hypophysis.

ADRENALS

A comparative study of the effect of adrenaline on the blood sugar, blood pressure, denervated heart, nictitating membrane, stomach, iris, hair and salivary gland in the cat. Chang, S., Chinese J. Physiol. 12:397. 1937.

The minimal amount of adrenalin that can cause activity under chloralose anesthesia was measured on different organs of a large number of cats. Salivary secretion was found in 13 animals to follow 0.011 mg. adrenaline per kilo. Hair movement occurred with 0.0048 mg./kg., using 17 cats. Dilation of the iris followed 0.003 mg./kg. (12 cats). Stomach relaxation was determined in 33 cats and followed 0.0021 mg./kg. The minimal amount of adrenaline causing contraction of the nictitating membrane was found to be 0.0017 mg./kg. in 30 cats. The acutely denervated heart was accelerated by 0.00075 mg./kg. (23 animals). Blood pressure rise in a cannulated carotid was controlled by injection of 1 cc. of saline. A series of dilutions of adrenaline, given to 41 cats, showed that an increase followed 0.00029 mg./kg. and that, contrary to Cannon and Lyman, small doses did not cause a depression of blood pressure. An increase in the fasting blood sugar of 7.8% in 15 min. was taken as a positive effect. This occurred after 0.000,0087 mg./kg., as tested in 21 animals ranging in weight from 1.3 to 3.0 kg. Each animal was tested with solutions of 1 to 1 million, 5 million, 10 million, 50 million and 100 million. Salivary secretion, dilation of pupil and blood pressure changes were observed in the same animals; hair erection, stomach relaxation and contraction of nictitating membrane in another group, and changes in the rate of the denervated heart in a third.—R. L. Zwemer.

Response to cold after reduction of adrenal tissue. Horvath, S. M., F. A. Hitchcock and F. A. Hartman, Am. J. Physiol. 121:178. 1938.

Studies of the respiratory metabolism of normal and unilaterally adrenalectomized albino rats at environmental temperatures of 29° and 4°C., and at 29° after exposure to 40°C. for periods of 2 to 72 hr. show that, *1)*, the basal heat production of albino rats is unchanged following the removal of one adrenal; *2)*, normal albino rats after exposure to an environmental temperature of 4°C. have an average increase in heat production measured at 29°C. of about 22%. The corresponding increase in heat production of unilaterally adrenalectomized animals is only about 7%; *3)*, at an environmental temperature of 4°C. the maximal heat production of unilateral adrenalectomized rats was attained more slowly and was lower than that of normal animals; *4)*, the colonic temperatures of normal and singly adrenalectomized rats remains practically unchanged at environmental temperatures of both 4° and 29°C.—Authors' Summary.

The effect of adrenalin in scorbutic animals (Über die wirkung des adrenalins bei C-hypovitaminotixchen tieren). Kasahara, M., N. Yoshito and H. Shigeo, Klin. Wchnschr. 16:1618. 1937.

In guinea pigs under urethane anesthesia, after scorbutic symptoms had appeared on a scorbutic diet, the intravenous injection of 0.001 mg. of adrenalin produced no effect on the arterial blood pressure. The simultaneous injection of 20 mg. of ascorbic acid with such a dose of adrenalin produced a marked rise in blood pressure, although the ascorbic acid alone had no vasopressor effect.—M. H. F.

ENDOCRINE GENERAL

Amenorrhea: its causation and treatment. Frank, R. T., M. A. Goldberger, U. J. Salmon and Gertrude Felshin, J.A.M.A. 109:1863. 1937.

For their study the authors selected patients who had suffered with amenorrhea for various periods and were of various ages. Intelligence, faithful carrying out of orders, ability and willingness to report as directed were of importance in the choice. In one instance the investigation covered 430 days. In only five cases were the studies continued for less than one entire month. By all available clinical criteria determined as a routine before selecting the patient, including the secondary sex characters, basal metabolism, blood examinations, sellar x-ray films, Janney test for sugar tolerance, blood pressure, pelvic examination, and the like, twenty-four of the twenty-seven patients showed no serious deviations from any group of normal women of similar economic and social status. A wide variation in the hormone status occurred. Evidence of almost complete ovarian afunction, subthreshold function and normal follicular activity as well as excessive activity was represented. In these four groups the gonadotropic assay may show either overfunction or underfunction. No evidence point-

ing to either a primary pituitary or a primary ovarian causation of amenorrhea could be demonstrated. Amenorrhea does not preclude the occurrence of ovulation or pregnancy. The response to estrogenic therapy of amenorrheic patients differs markedly from that in the menopause. The threshold of response in amenorrhea is far higher than in the menopause. This difference can be utilized in patients to differentiate between the two conditions, if an excess of gonadotropic substance has been found in the urine. Disappearance of gonadotropic substance produced by 30,000 rat units of estrogenic substance warrants the diagnosis of menopause. No useful purpose is served in prescribing estrogens for the treatment of amenorrhea. In the dosage (from 60 to 510 rat units) used by the authors, gonadotropic preparations likewise proved ineffective. It is justifiable to try very high dosage of gonadotropic preparations when they become available. The study has failed to locate the cause or causes producing amenorrhea. Not only ovarian or anterior pituitary refractoriness but also a failure of uterine response must be considered in the etiology.—Abst., A.M.A. Service.

Effects of acetylcholine and adrenalin on nictitating membrane. Morison, R. S. and G. H. Acheson, Am. J. Physiol. **121**:149. 1938.

The variation of the response of the nictitating membrane of the cat with the doses of adrenalin and acetylcholine injected into the blood stream is expressed by a hyperbola. Both substances produce the same maximal contraction. The responses to simultaneous applications of the two drugs are not the linear sum of the responses due to each of the drugs. Adrenalin increases the effect of acetylcholine as much as an equivalent amount of acetylcholine. This result presents difficulties to the view, expressed by Rosenblueth, that the action of these drugs is due to a monomolecular reaction with a single cell receptor.—E. Bozler.

GONADS

Hormonal causes of prolonged gestation (Ueber die hormonalen Ursachen der Uebertragung). Arvay, A. V., Zentralbl. f. Gynäk. **61**:2900. 1937.

The author attempts to decide whether the corpus luteum is essential to that action of gonadotropic materials shown by reducing the sensitivity to oxytocic extracts of the posterior pituitary. The tests are made in rabbits by prolonging gestation beyond some 30 days, sometimes to as much as 60 days. This can be done with progestin, with pregnancy serum preparations, still better when pregnancy serum is ether extracted to remove estrin, or with similarly treated urine from pregnant women. It seems improbable that this action is via stimulation of corpus luteum function, because the delay in parturition can be effected even though the therapy begins only when signs of delivery are appearing. Since attempts to remove ovaries from animals during gestation precipitated abortion, the author used irradiation to destroy ovaries. Tissue studies convinced him of the destruction, and he found no corpora lutea. Yet therapy prolonged gestation. Further experiments showed that with large doses of estrin abortion could be induced. On surviving uteri, anterior pituitary treatment eliminated the response to posterior lobe extracts in the bath. The death of the fetus in prolonged gestation is held to be due to the time factor, not to the toxicity of gonadotropic material, since a period of therapy interrupted at term did not kill the fetus. Based on these results, the use of gonadotropic substance is recommended for threatened abortion, and of estrin for weak labor pains.—E. L. S.

The general significance of the disorders of menstrual interval and flow. Burch, J. C., Internat. Clin. **4**:(series 47)65. 1937.

Menstrual disorders, in general, consist of 2 great classes. The first of these consists of the organic diseases affecting menstruation, such as cancer, fibroids, ectopic gestation, abortions, polyps, etc. In the second class there are symptoms of pain or bleeding, or disordered interval without any of the organic causes being present. Disorders of interval and flow are the results of the following causes: *a)*, general constitutional diseases depressing the endocrine system, such as tuberculosis, focal infections, nutritional disturbances and many others; *b)*, diseases of the thyroid gland, the majority of which can be classified as hypothyroidism; *c)*, diseases of the pituitary gland, most common of which is pituitary dysfunction and the next most common, pituitary failure; *d)*, diseases of other endocrine glands, such as the adrenal and pancreas, which are very rare contributors to the production of disorders of interval and flow; *e)*, diseases of the ovary, such as are the result of surgical operations, destruction by inflammatory diseases, or developmental failure. Changes in the menstrual interval and flow are among the earliest of symptoms indicating constitutional or endocrine diseases; such changes should always be considered both from the constitutional and local aspect; from the constitutional standpoint, all patients with these diseases should have a very thorough and exhaustive study.—I. B.

Total urinary estrogen, estrone and estriol during a menstrual cycle and a pregnancy. Smith, G. V. S., O. W. Smith and G. Pincus, Am. J. Physiol. 121:98. 1938.

In following the excretion of 'total' estrin and of estriol and estrone separately, two methods of liberating the estrin from its conjugated form were compared. The estrogenic fractions were bioassayed; simultaneous colorimetric estimations were also conducted for comparative purposes. Two peak levels of estrogen excretion were noted in the menstrual cycle of 25 days: the first during the 9th-11th days, and the second on the 15th-23rd days of the cycle. Very low levels of estrin were present in the urine during menstrual flow. Although more estrone than estriol was excreted throughout the cycle, the estriol maintained a relatively high level during the 9th-23rd days. The authors associate this with the luteal phase of the cycle, when a higher level of progestin is present. The excretion of total estrin during the gestation increased gradually, reaching a peak level 2 weeks before delivery, after which a marked drop occurred. The estriol excretion increased more rapidly than that of estrone during the pregnancy, and reached a ratio of 15 at 9 months and 8 days. This ratio change is explained as due to progestin, probably formed in large amounts in the placenta. Samples assayed after the 112th day of term gave considerably higher value by bioassay than by the colorimetric method. The authors interpret this as indicating the presence in late pregnancy urine of a third estrogen which is biologically more active and/or is less chromagenic than either estriol or estrone (e.g., estradiol).—S. L. Cohen.

Ketonic and non-ketonic estrogens. Westerfeld, W. W., and E. A. Doisy, Ann. Int. Med. 11:267. 1937.

Using Girard's reagent and semicarbazid, the authors were able to separate quantitatively the ketonic from the non-ketonic estrogens. Ketonic estrogens were found in sow ovaries and human placenta but none in cow ovaries. By determining the relative amounts of two forms excreted after injection into normal and castrate-hysterectomized monkeys it was shown that the reaction between theelin and dihydrotheelin is reversible, and that the ovaries and uterus are not essential for the transformation.—R. R. Durant.

HYPOPHYSIS

Hypophysectomy and replacement therapy in relation to the growth and secretory activity of the mammary gland. Gomez, E. T., and C. W. Turner, Univ. Missouri Agr. Exper. Sta. Bull. No. 259. 1937.

Replacement therapy with ovarian hormones alone and in combination with various purified pituitary hormones, and desiccated whole sheep pituitaries was ineffective in stimulating the growth of glandular parenchyma of the mammary gland of hypophysectomized male and female laboratory mammals. The implants of adult normal male rat pituitaries were likewise ineffective. Twenty to 25 daily implants of 1 adult male rat pituitary obtained from donors which had previously been injected with estrogen daily for 10 to 20 days stimulated complete hyperplasia of the mammary gland parenchyma of normal and castrated-hypophysectomized male and female guinea-pigs. Hypophysectomy of lactating guinea-pigs and cats at any time during the lactation period was immediately followed by a rapid cessation of milk secretion. Unlike the crude pituitary extracts, the replacement therapy with galactin alone and in combination with thyroxine or thyrotropic hormone beginning immediately after hypophysectomy or after the complete cessation of lactation after the operation, did not prevent the rapid cessation or re-establish lactation. The injection of eschatin or adrenotropic hormone alone was ineffective in stimulating the secretion of milk. The simultaneous injection of galactin, eschatin or the adrenotropic hormone and a glucose solution prevented the cessation of lactation after hypophysectomy in the guinea-pigs, and by continued treatment lactation was maintained for periods comparable to that observed in normal parturient animals. The above factors are indispensable for the initiation and maintenance of lactation. In the light of the results presented, a discussion of the endocrine control of the mammary gland is also included.—Authors' Abstract.

The relation of the hypophysis to changes in sugar tolerance and insulin sensitivity induced by changes of diet. Himsworth, H. P., and D. B. McN. Scott, J. Physiol. 91:447. 1938.

When normal rabbits are fed a high carbohydrate diet, they have an increased sugar tolerance and insulin sensitivity, whereas a low carbohydrate diet impairs the sugar tolerance and lowers the sensitivity to insulin. Eight 2-kg. male rabbits survived hypophysectomy and the experimental dietary procedure, and were found to be insulin-sensitive with a high sugar

tolerance regardless of diet. However, treatment of the hypophysectomized rabbits with Young's hypophyseal glyotropic factor lowered the sugar tolerance and decreased the insulin sensitivity.—R. L. Zwemer.

The lactogenic and thyrotropic hormone content of the anterior lobe of the pituitary gland. Reece, R. P., and C. W. Turner, Univ. Missouri Agr. Exper. Sta Bull. No. 266. 1937.

An assay method was developed for the determination of the minute quantities of the lactogenic hormone in pituitary glands from laboratory animals. An extensive study was made of the rat pituitary gland. As the male rate matured there was an increase in the lactogen content per pituitary gland, due entirely to increased pituitary size. In the female there was an increase in lactogen concentration in the pituitary gland with the onset of sexual maturity. During pregnancy there was no increase in the lactogen content whereas following parturition the quantity of the hormone was doubled. Suckling, either with or without the removal of milk, decreased the lactogen content of the pituitary gland. Castration did not alter the lactogen content of the pituitary gland, but ovariectomy decreased the content. The abdominal anchorage of the testes did not alter the lactogenic hormone content of the pituitary gland. Estrogen injections into ovariectomized rats, normal and castrated male rats, and normal male guinea pigs increased the lactogen content per pituitary gland while progestin injections into ovariectomized rats had no influence upon the pituitary lactogen content. Attempts were made to inhibit lactation with ovarian hormone injections. The lactogen content of bovine fetal pituitaries was the lowest found in any of the cattle pituitary assays. Pituitaries from heifers contained more lactogen than glands from bulls. As the heifers matured there was an increase in the hormone concentration in the pituitary gland, but as the bulls matured the hormone concentration remained unchanged. Pituitary glands from bulls contained more lactogen than glands from steers. Pituitary glands from dairy cows contained larger quantities of the lactogenic hormone than glands from beef cows. The thyrotropic content of pituitary glands was determined in the growing and in the mature animal. Pituitaries from bulls contained more of the thyrotropic hormone than glands from heifers and heifer pituitaries contained more than glands from steers. Pituitaries from dairy cows contained more of the thyrotropic principle than glands from beef cows.—Authors' Summary.

Pituitary as probable factor in origin of headaches of menopause. Reel, P. J., and T. F. Lewis, Ohio State M. J. 33:156. 1937. Abst., Arch. Neurol. & Psychiat. 39:619.

Pituitary headaches may be entirely occipital or suboccipital in type. Some patients complain of occipital pain transmitted bilaterally to the frontal or temporal areas. Quite commonly one entire side of the head is involved. Dull, heavy distress located behind the inner angles of the eyes is frequently encountered. The pain, regardless of type, is usually severe. It may be lancinating, or may develop as a deep-seated, heavy, dull ache. In 14 of the 19 patients, an abnormal amount of gonadotropic substances was demonstrated in the blood prior to irradiation, according to the Fluhmann technic. Roentgen treatments were given with 200 kilovolts at a distance of 50 cm. with 0.65 mm. of copper and 1 mm. of aluminum as a filter and an 8 cm. mask. A total of 1,000 r was used, divided into two exposures of 500 r each, administered a week apart and on opposite sides of the head. Other than temporary alopecia in 2 patients, and slight transient parotitis in 1, no complications or ill effects were encountered. After irradiation, 14 of the 19 patients showed a marked degree of improvement. Three of the remaining 5 were improved to some extent, whereas the remaining 2 received no benefit. The authors believe that the total dose has an inhibitory effect on the activity of the hypophysis with diminution in the size of the gland and subsequent lessening of tension within the sella. Since headache tends to disappear under this management without the aid of other forms of therapy, the pituitary is a probable factor in the causation of certain types of headache peculiar to the menopause.—(Abbreviated)

The 'pancreatropic' action of anterior pituitary extracts. Richardson, K. C., and F. G. Young, J. Physiol. 91:352. 1937.

The action of anterior pituitary preparations on pancreas islets and on blood sugar was studied. Schockaert's 1931 type A.P. extract, Robertson's 'tethelin' and a commercial preparation made by Mulford were used. All were adjusted so that 2 cc. = 1 gm. fresh gland. Controls received non-specific tissue extract. Wistar rats on Wistar diet were injected with 2 cc. daily from 3 days to 3 weeks. The pancreas from a sacrificed animal was given special histologic treatment, and the islet x100/acinar ratio and absolute acinar changes were determined by the paper tracing and weighing technic. In 10 controls the range was 0.9 to 1.5, with a mean of

1.1 ± 0.06. An equal number given fresh saline extract of A.P. were from 0.75 to 4.5 with a mean of 2.2 ± 0.35. Saline extracts of acetone dried glands showed a ratio of 1.5 ± 0.20 in 10 cases. In 2 of 3 rats given saline extract of ether-soluble fraction of fresh glands, a significant increase occurred. The rats receiving the alcohol-soluble part of fresh glands and those with 'tethelin' were not significantly affected. In the above experiments the finding of giant cells was not correlated with islet increase, nor was there a striking rise or fall in the 62 blood sugars determined at various intervals after injection.—R. L. Zwemer.

The relation of the hypophysis and adrenal cortex to the removal of excess glucose from the blood of rats. Samuels, L. T., H. F. Schott and H. A. Ball, Am. J. Physiol. **120**:649. 1937.

This study was carried out to determine whether the decreased sugar tolerance of hypophysectomized rats was a direct or indirect effect of the gland removal. Approximately 120 male rats 4-6 months old were used, 17 and 21 days after hypophysectomy, and 1-28 days after adrenalectomy in the case of operated animals. Blood sugar determinations were made by the micro-method of Folin. The decreased ability of hypophysectomized rats to remove excess glucose from the blood is not primarily dependent on the adrenal cortex, fasting, circulatory depression or urinary loss. Although not influencing the glucose tolerance, removal of adrenals in hypophysectomized rats introduces further disturbances in metabolism, which decrease the length of life on fasting considerably below that of either the hypophysectomized or the bilaterally adrenalectomized animals. The metabolic influence of the pituitary cannot then be solely through the adrenal. The rate of glucose storage appears to be much reduced in hypophysectomized rats. Tissue utilization of glucose seems to be increased in the rat without pituitary.—K. Brownell.

The adrenals and gonads of rats following thyroidectomy considered in relation to pituitary histology. Zeckwer, I. T., Am. J. Physiol. **121**:224. 1938.

White rats were thyroidectomized at 24 to 88 days of age, and were killed 76 to 356 days after operation. The changes observed in the pituitaries were loss of acidophiles and accumulation of thyroidectomy cells, which differ from castration cells. The adrenal and gonad weights were regarded as measures of the adrenotropic and gonadotropic activity of such pituitaries. Kidney weight was taken as an index of visceral growth. In males there was no significant change in weights of adrenals and testes, but kidney growth was retarded. In females the adrenal and gonad weights were decreased in proportion to retardation of kidney growth. The author regards his results as evidence that adrenal growth does not depend on acidophiles or thyroidectomy cells, but that it may depend on a basophile cell.—C. Duncan.

PANCREAS

Myxedema and diabetes mellitus: A case report with autopsy findings. Carey, J. B., S. L. Arey and E. H. Norris, Ann. Int. Med. **11**:838. 1937.

A report of the detailed clinical history of 15 years' duration of a case of myxedema upon which was superimposed diabetes mellitus terminating with intestinal obstruction from carcinoma of the sigmoid. The effect of thyroid therapy on carbohydrate metabolism is stressed.—R. R. Durant.

The effect of hexylresorcinol upon the absorption of insulin from the gastro-intestinal tract of dogs. Daggs, R. G., W. R. Murlin and J. R. Murlin, Am. J. Physiol. **120**:744. 1937.

The purpose of the work was to investigate the effect of insulin, administered orally in conjunction with small amounts of hexylresorcinol and Na_2CO_3 on the blood sugar of normal dogs. Basal blood samples were taken from 16-hr.-fasted dogs, after which various quantities of Na_2CO_3, insulin and hexylresorcinol were given by stomach tube and the blood sugar curve followed for 2-3 hr. thereafter. Benedict's method was used for sugar determinations. A blood sugar drop of over 20 mg.% was considered significant. In about 50% of the experiments this treatment produced a significant, although not prolonged, drop in blood sugar.—K. Brownell.

Factors influencing the prognosis in diabetic coma. Dillon, E. S., and W. W. Dyer, Ann. Int. Med. **11**:602. 1937.

A study of the clinical and laboratory records of 3009 cases of diabetes in 224 individuals was undertaken in order to discover the more important factors influencing the prognosis in

diabetic coma, and to explain the higher incidence of acidosis among the lower economic classes. To do this, it was necessary to classify the cases as to the type and severity of complications, some of which were not recognized until postmortem examination. The most significant factors were the type and severity of complication, the blood urea nitrogen, advancing age, colored race, the mental state (conscious or unconscious) and the duration of the acidosis. Factors which proved to be much less significant are blood sugar, CO_2 combining power and leucocytosis. The authors conclude that a much more accurate prognosis can be given from the clinical data than from the laboratory data.—R. R. Durant.

Induced hyperinsulinism. Dombrowski, E. F., H. H. Goldstein, A. P. Bay and J. V. Adlin, Illinois M. J. **73**:147. 1938.

Patients given insulin shock treatment were studied to observe changes produced by induced hypoglycemia. It was noted that symptoms could be grouped under 5 headings: respiratory, vasomotor, neurologic and mental, cardiovascular and temperature changes. Each group is deserving of a complete and separate study, which will be undertaken by the authors.—I. B.

Treatment of diabetes mellitus with insoluble insulin compounds. II. Histone-insulin. Gray, P. A., F. Bischoff and W. D. Sansum, Ann. Int. Med. **11**:274. 1937.

The treatment of 30 diabetic patients with histone-insulin for periods as long as 3 months shows it to be comparable to protamine zinc insulin in action. However, no allergic reactions or untoward local reactions have been experienced in its use. The average dosage required is about the same as that of regular insulin.—R. R. Durant.

Absorption of insulin from Thiry-Vella loops of the intestine in normal and depancreatized dogs. Murlin, J. R., Ruth L. Tomboulian and H. B. Pierce, Am. J. Physiol. **120**:733. 1937.

The absorption of insulin from Thiry-Vella loops of the jejunum, as judged by reduction in blood sugar, was studied in 4 dogs while normal, and also in 2 after pancreatectomy. Positive evidence of absorption was obtained in 61% of the 80 trials with normal dogs and in 73% of the 14 trials with the depancreatized. It was found that several different substances put into the loop as a preparatory wash before the introduction of insulin favored its absorption. The most successful were 0.1% HCl, bile and hexylresorcinol. It is probable that bile and hexylresorcinol act in virtue of their surface-lowering effects. The most favorable medium in which to dissolve the insulin for presentation to the mucosal surface proved to be 1/20-1/40N $NaHCO_3$. There was no very close relation between the concentration of insulin and the effect. The least concentration to produce a marked reduction of blood sugar was 35 clinical units in 10 cc. Doses of over 100 U. in this volume gave somewhat larger effects as a rule than less than 100. The concentration of hexylresorcinol necessary to produce a favorable effect was 0.1-0.5%. Insulin can be inactivated by exposure to intestinal juice at body temperature for 20-30 hr. Whether or not this is a true digestive destruction has not been determined.—K. Brownell.

Fetal and maternal mortality in diabetes. Potter, E. L., and F. L. Adair, Am. J. Obst. & Gynec. **35**:256. 1938.

Besides offering a careful review of the literature from the past 10 years, the authors present brief records about 16 pregnancies in 15 diabetic women seen over 5 years, approximately 1 in 900 deliveries. The usual increased risk of toxemia, etc., is confirmed, but the fetal mortality of 31% and maternal mortality of only 1 case compares very favorably with other data published from the insulin era.—E. L. S.

Problems connected with the use of protamine-zinc-insulin. Ricketts, H. T., Ann. Int. Med. **11**:777. 1937.

A report of clinical experience with protamine-zinc-insulin is given in 26 cases of diabetes. Severe cases were found more difficult to manage on protamine-zinc-insulin; however, carbohydrate diets were found more satisfactory, and a bed-time meal was required in two-thirds of the cases. Some cases required 5-15 U. of regular insulin given with the protamine-zinc-insulin, to prevent glycosuria during the day at the same time avoiding hypoglycemia in the morning.—R. R. Durant.

A relationship between the blood sugar level and the rate of sugar utilization, affecting the the theories of diabetes. Soskin, S., and R. Levine, Am. J. Physiol. **120**:761. 1937.

These experiments were undertaken in an attempt to reconcile the non-utilization and

the over-production theories of diabetes. Fifteen normal and 12 depancreatized dogs were used. All dogs were fasted 3 days. When pancreatectomy was performed it was done on the 1st day of fast and no insulin administered. On the fourth day aviceration was performed, the ureters tied off and intravenous dextrose injection instituted with the aid of a constant injection pump, the rate being determined by the blood-sugar level desired. Sugar utilization was determined from lactic acid, muscle glycogen determinations made at the beginning and end of a 4-hr. period, and blood sugar determinations made at half-hour intervals over the same period. The completely depancreatized dog in the absence of insulin can utilize dextrose at any rate of which the normal animal is capable. In both the normal and depancreatized dog, the rate of dextrose utilization depends upon the height of the blood-sugar level. Within a certain glycemic range, the depancreatized dog utilizes less sugar at any given blood-sugar level than the normal animal at the same level. But the depancreatized dog, at its normal hyperglycemic level, utilizes as much or more sugar than the normal dog at its usual normal blood-sugar level. On this basis, the two supposedly opposing theories of diabetes may be reconciled. The non-utilization theory applies only in a particular restricted sense. The over-production theory remains as the major component of the new conception.—K. Brownell.

Studies on the constitution of insulin. III. The acetylation of insulin by ketene. Stern, K. G., and A. White. J. Biol. Chem., **122**:371. 1938.

Insulin was treated with ketene ($CH_2 = C = O$) at a pH of 5.7 for 5 minutes at room temperature to acetylate the free amino groups. A prolonged treatment for 30 hours or more was employed to acetylate the hydroxyl groups of tyrosine. The activity of the preparations in which only the amino groups were bound was not appreciably affected. When the hydroxyl groups of the tyrosine part of the insulin were bound, the pharmacodynamic action of the insulin decreased as the degree of acetylation increased. It was concluded that the free amino groups were dispensable, but the tyrosine hydroxyl groups were indispensable for the physiological activity of the hormone.—J. M. L.

Clinical investigations of insulins with prolonged activity. Wilder, R. M., Ann. Int. Med. **11**:13. 1937.

Clinical evidence is presented showing the effect of calcium and zinc, in respectively diminishing and extending the time of action of protamine insulin. Protamine insulin was found to produce hypoglycemia gradually, without the warning signs characteristic of unmodified insulin, therefore necessitating greater attention to the diet. The duration of action of crystalline insulin was found to be intermediate between that of unmodified and protamine insulin. The use of protamine insulin demanded lower carbohydrate diets, although some cases were found to be insensitive to protamine insulin, therefore permitting the use of more carbohydrate. The necessity of continuous protection of body proteins by insulin was demonstrated. The author believes that protamine insulin has a distinct advantage when used with regular insulin in the treatment of diabetic acidosis.—R. R. Durant.

PARATHYROIDS

Changes in the teeth following parathyroidectomy. I. The effects of different periods of survival, fasting and repeated pregnancies and lactations on the incisor of the rat. Schour, I., S. B. Chandler and W. R. Tweedy, Am. J. Path. **13**:945. 1937.

The effect of parathyroidectomy on the incisor of the albino rat was studied in 100 rats (3 to 360 days after operation) and 27 controls, in respect to gross changes (including roentgenograms and eruption rates) and microscopic alterations. The incisor of the parathyroidectomized rat shows disturbances in calcification and in development. The essential character of the reaction varies with the length of the survival period. In short survivals up to 20 days the postoperative dentin indicates denser calcification than normal, and is demarcated from the pre-operative dentin by a distinct ring, which stains deeply with hematoxylin. This ring, which is formed at the time of the operation, and which is also found in other related experimental conditions, appears to be an acute response to a shock to calcium metabolism, and may be referred to as the calciotraumatic ring. The denser calcification of the postoperative dentin in this group may be a result of the calcium retention reported to persist during the early survival period. In longer survivals, the changes consist of defective calcification and defective formation of enamel and dentin. The severity of the changes increases with the increase in time of survival. The most characteristic change consists of an aberration in dentin

calcification. The dentin shows an irregular and accentuated alternation of zones of different densities in calcification. This severe fluctuation, rather than lack of calcification, is responsible for the readiness of the parathyroprivic dentin to fracture. The histological changes are aggravated by fasting on every seventh day. The most severe alterations were found in animals subjected to repeated pregnancies and lactations. Histological examination showed no evidence of calcium withdrawal from the calcified tissues of the teeth.—W. J. A.

Changes in the teeth following parathyroidectomy. II. The effect of parathyroid extract and calciferol on the incisor of the rat. Schour, I., W. R. Tweedy, S. B. Chandler and M. B. Engel, Am. J. Path. 13:971. 1937.

This study was based on 59 parathyroidectomized rats that were operated on between the ages of 21 and 93 days. The animals were divided into 2 groups. In the first group 29 received 1 to 4 injections of parathyroid extract. The total dosage ranged from 10 to 274 (Collip) U. The survival period was from 3 to 17 days. Of the second group 30 received a single administration of 46,000 to 644,000 I.U. of calciferol. The survival period was from 1 to 10 days. Serum calcium values were obtained immediately preceding death.

The histological findings in the dentin of the incisors of these animals were similar in the first and second groups, and consisted of *a*, a primary response characterized by an eosin staining zone; *b*, a secondary response in the form of a hematoxylin staining zone; *c*, in the majority of cases a sharp hematoxylin staining ring at the border of the pre-experimental and postexperimental dentin; and *d*, cytological changes in the active enamel-forming cells in one-half of the animals.

In the second group the serum calcium was found to rise during the first 3 post-administration days, and then to decline more slowly to the original level. The level and rapidity of the rise increased with the dosage. The serum calcium values were lower than those found in corresponding conditions in the unoperated animals. The histological effect of single doses lasted as long as 10 days in animals treated with parathyroid extract and calciferol. In 2 rats treated with parathyroid extract the effect lasted 17 days.—W. J. A.

PITUITARY

The effect of water deprivation on the fluid exchange of cats with diabetes insipidus. Fisher, C., H. W. Magoun and A. Hetherington, Am. J. Physiol. **121**:112. 1938.

The effect of water restriction on 20 cats with diabetes insipidus experimentally induced by use of the Horsley-Clarke stereotaxic instrument was studied over a period of weeks. The results indicate that the polyuria of diabetes insipidus is primary, and agree with the previous report of Fisher, Ingram and Ranson (1938) and that of Richter (1935) who used rats. The polyuria developed in the latent period, even though the water intake was restricted and a negative water balance developed. This negative balance was compensated when the cats were given free access to water. If the fluid intake was restricted to a level below the preoperative level, the diabetic cats in some cases did not survive, even though normal cats suffered no ill effects when receiving the same treatment. In giving the water to diabetic cats by stomach tube, it was clearly demonstrated that the mucosa of the buccal cavity did not have to be moistened, and that thirst was dependent on bodily hydration.—Lena Lewis.

THYROID

A study of the effect of vitamin B and iodine on the weight, iodine content and structure of the thyroid gland of the rat. Carpenter, Mary D., and G. Sharpless, J. Nutrition 13: 235. 1937.

It was found that vitamin B deficiency alone did not affect the size, iodine content, or structure of the thyroid, but rats fed a vitamin B deficient diet containing 0.0038% iodine had thyroids with a structure simulating colloid goiter. Vitamin B or a higher iodine intake prevented the condition. The results obtained with other diets suggest that some factor is lost in autoclaved yeast and not supplied by a vitamin B containing extract of wheat germ which causes an increase in the thyroid iodine. The iodine content of the glands varied from 4 γ in animals fed diets containing no added iodine and low in vitamin B to 32 γ in those fed diets containing both iodine and vitamin B. The iodine content of the glands was low with low food intake regardless of iodine intake. The weight of the hypophysis was not significantly affected by vitamin B and iodine variations.—Author's Abst., Wistar Bib. Service.

Diagnosis of hyperthyroidism masquerading as heart disease. Ervin, C. E., and R. F. Dickey, Pennsylvania M. J. 41:506. 1938.

In simple hyperthyroidism the associated tachycardia is not appreciably reduced by rest alone nor by digitalization, but rather by the use of iodides. In heart disease not associated with toxic goiter, the tachycardia is not influenced by the iodides but by rest and the proper use of digitalis. When a supposedly heart disease tachycardia does not respond to rest and digitalization, one of 2 situations exists: the heart disease is complicated by thyroid toxicity, or digitalization is incomplete. Because of the almost specific responses to rest, to digitalis, and to iodides, their use individually and in combination is advocated, not only as therapeutic measures but as diagnostic procedures.—I. B.

The basal metabolic rate in normal pregnancy. Hanna, G. C., Am. J. Obst. & Gynec. 35: 155. 1938.

In a group of 57 women, basal rates were determined in the fifth and ninth months, and on the ninth day post partum. The average woman in the fifth month was about 8% above normal, in the ninth month 14 to 16% above normal, but post partum she was back to 3 to 4% above normal. (No data about the extent of variations or the prediction standards employed are published.)—E. L. S.

Some of the atypical manifestations of hyperthyroidism which obscure its diagnosis, with observations on some of the cardiac features of this condition. McMillan, T. M., and M. Wendkos, Internat. Clin. 3:213(Series 47). 1937.

Four cases of hyperthyroidism are presented and discussed. In 3 the diagnosis was obscured because of atypical manifestations and because of the domination of cardiac derangement. The fourth was a case of angina pectoris in a thyrotoxic woman of 25. The atypical forms of the first 3 cases were *a)* low grade chronic thyrotoxicos with constant variation in weight and with auricular fibrillation; *b)* apathetic hyperthyroidism, the usual nervous manifestations of the disease being replaced by apathy, and *c)* hyperthyroidism in an old man in whom the thyrotoxic manifestations might easily have been assigned to the degenerative changes of senility.—I. B.

Nutritional factors in Graves's disease. Means, J. H., S. Hertz and J. Lerman, Ann. Int. Med. 11:429. 1937.

Clinical evidence is presented showing the etiological rôle of malnutrition in Graves's disease. The deficiency may be in total caloric value or in one or more factors such as iron, calcium and vitamin A and B_1. The possibility of thyrotoxicosis following rapid weight reduction with or without the use of thyroid hormone is stressed.—R. R. Durant.

Lanugo and hypothyroidism. Infantile hirsutism [Lanugo et hypothyroidisme. Infantilisme pileux (Persistance du lanugo foetal) chez un enfant hypothyroidien, âgé de huit ans. Sa disparition á la suite du traitement par la thyroidine]. Mussio-Fournier, J. C., and J. M. Cervino, Bull. Acad. méd. 117:669. 1937.

The fetal hair disappeared after 2½ months treatment, and the mentality and other signs of thyroid deficiency showed improvement also.—A. E. Meyer.

Thyroidine and points of ossification (Thyroidine et points d'ossification. A propos d'une idiote mongolique, âgée d'un an, et traitée par la thyroidine, qui presentait des points d'ossification à l'âge de trois à quatre ans). Mussio-Fournier, J. C., and J. M. Cervino, Bull. Acad. de méd., Paris 117:666. 1937.

At the age of 4 months, treatment was initiated with a dose of 0.05 gm. daily, increasing gradually to 0.4 gm. at one year. A premature ossification was observed corresponding generally to the development of 3-4 years. The centers of ossification, however, showed considerable irregularity, some being far advanced whereas others showed no sign of stimulation.—A. E. Meyer.

Exophthalmic goiter and gastro-duodenal ulcer—two constitutionally different diseases. Robinson, S. C., Illinois M. J. 73:210. 1938.

The constitutional and etiological aspects of Graves's disease and of gastro-duodenal ulcer are compared and differentiated. While both conditions are psychogenic, the patients differ in

psyche and soma. The build of the ulcer patient is long and thin, while the hyperthyroid patient may be of any build but there is an inclination toward being slender and underweight. The 2 diseases seldom occur simultaneously in the same person. Ten males to 1 female have ulcer; 10 females to 1 male have exophthalmic goiter. The goiter patient tends toward achlorhydria; the findings in the ulcer patient are the direct opposite. The hyperthyroid patient presents a thymolymphatic constitution and sympathicotonia. This is absent in the ulcer patient. Other differential features are discussed in this paper.—I. B.

Hyperthyroidism in the negro. Porter, W. B., and H. Walker. Ann. Int. Med. 11:618. 1937.

A study of the hospital records of 37 negro patients suffering from Graves's disease shows this condition to be as common among negroes as whites in the geographical area of Richmond, Va., and environs. The only difference in reaction of the negro and white patient to hyperthyroidism is quantitative rather than qualitative. The high incidence in a non-endemic district leads to the conclusion that there must be etiological factors other than primary iodine deficiency or stress of urban life.—R. R. Durant.